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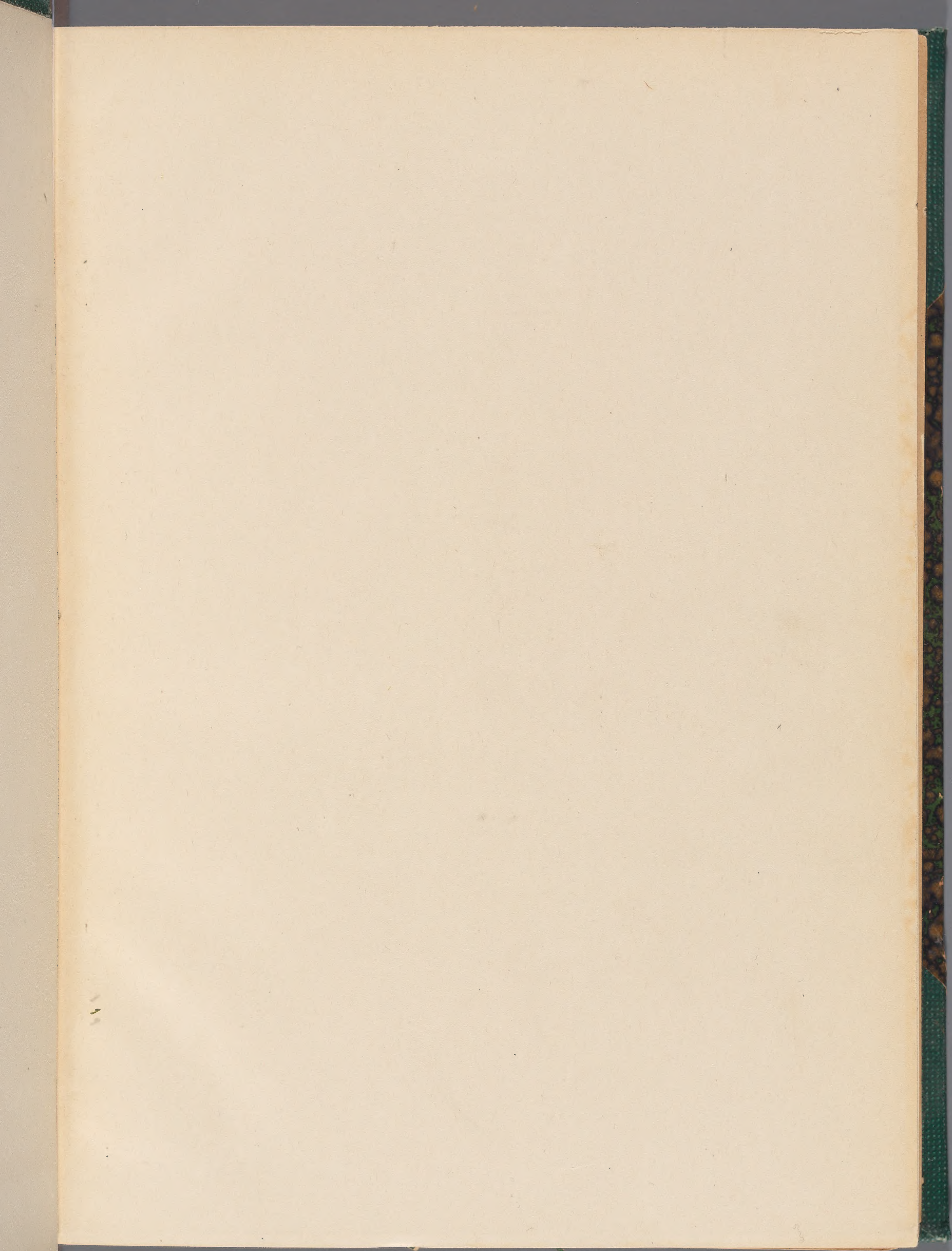
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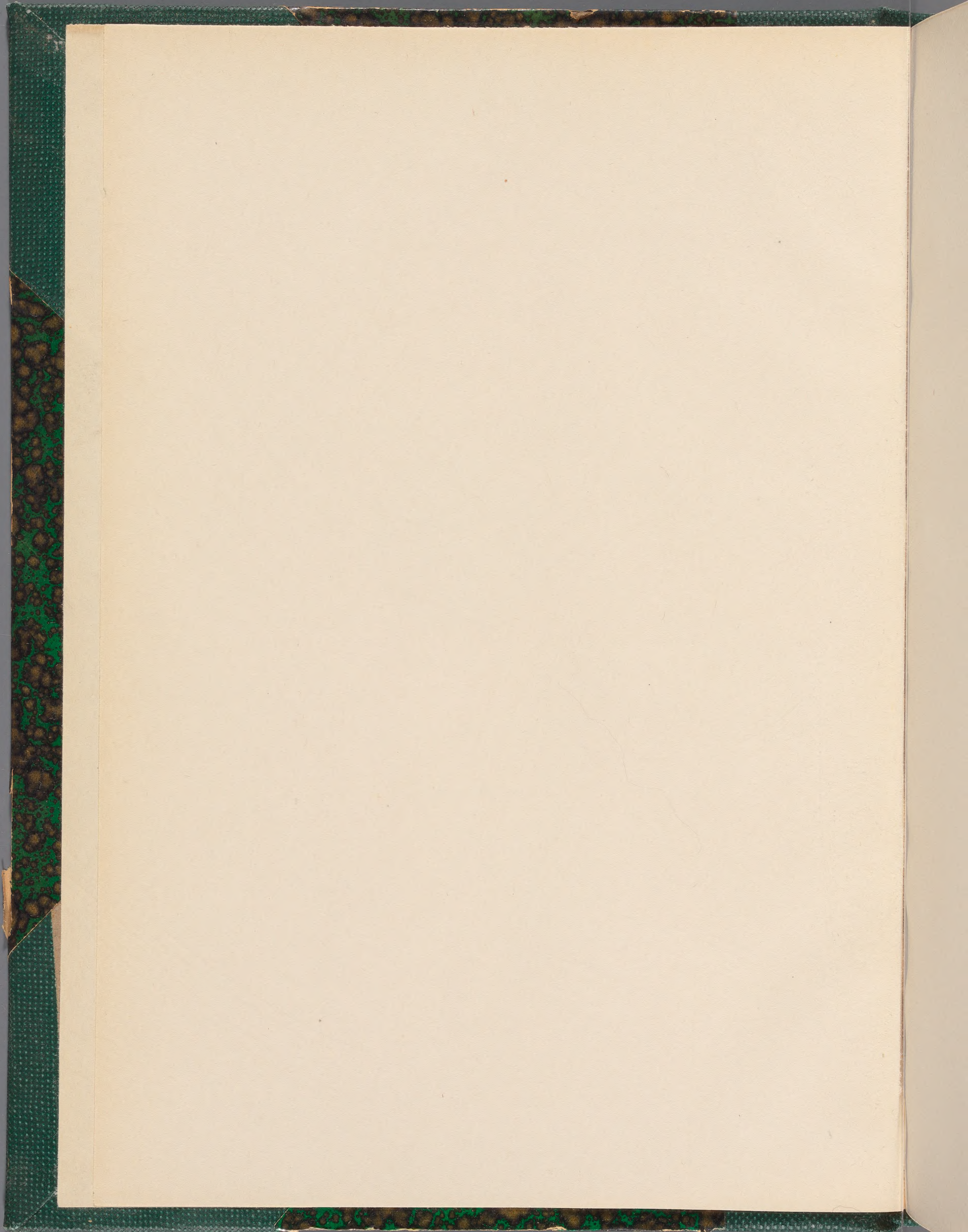
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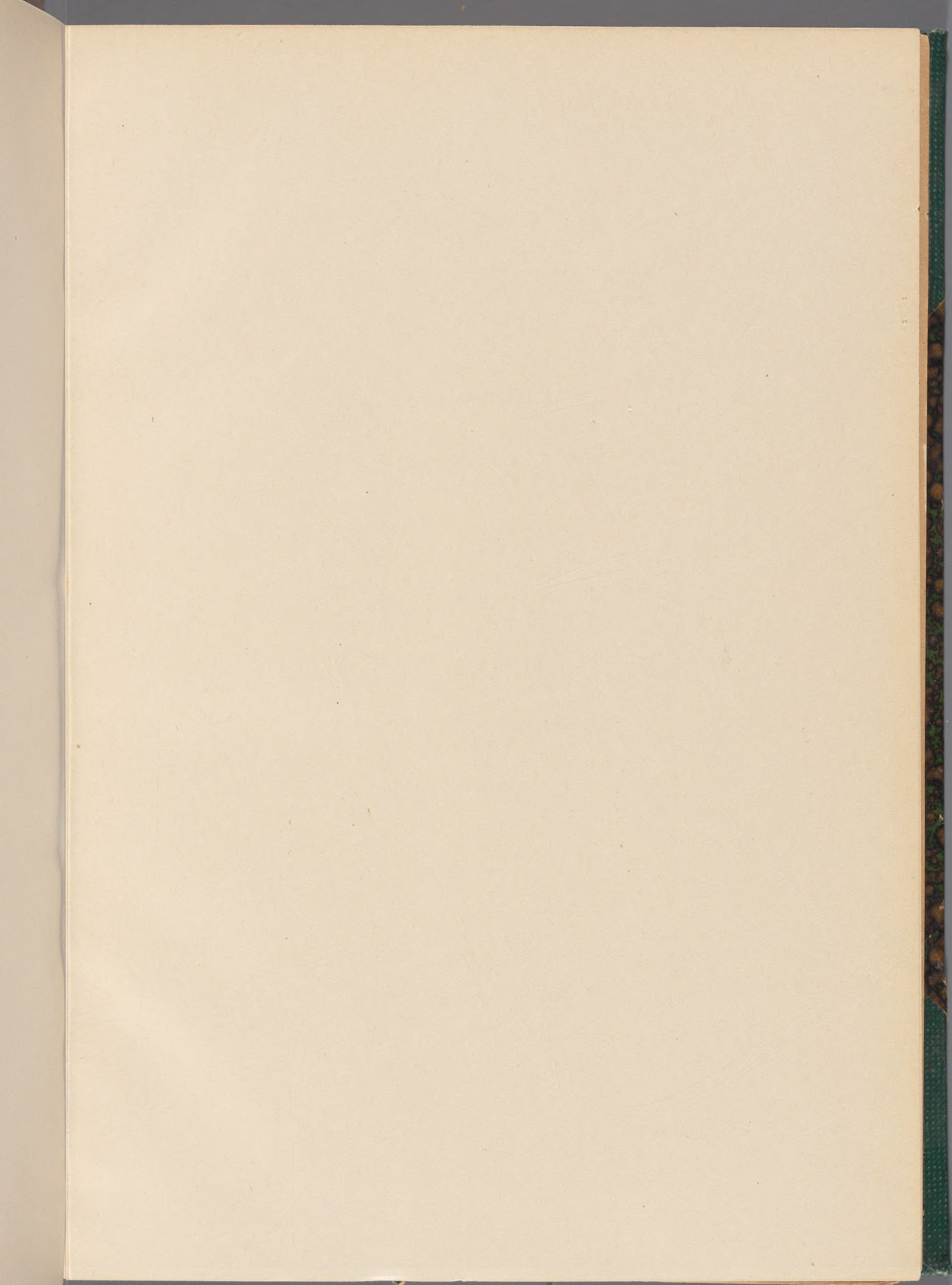
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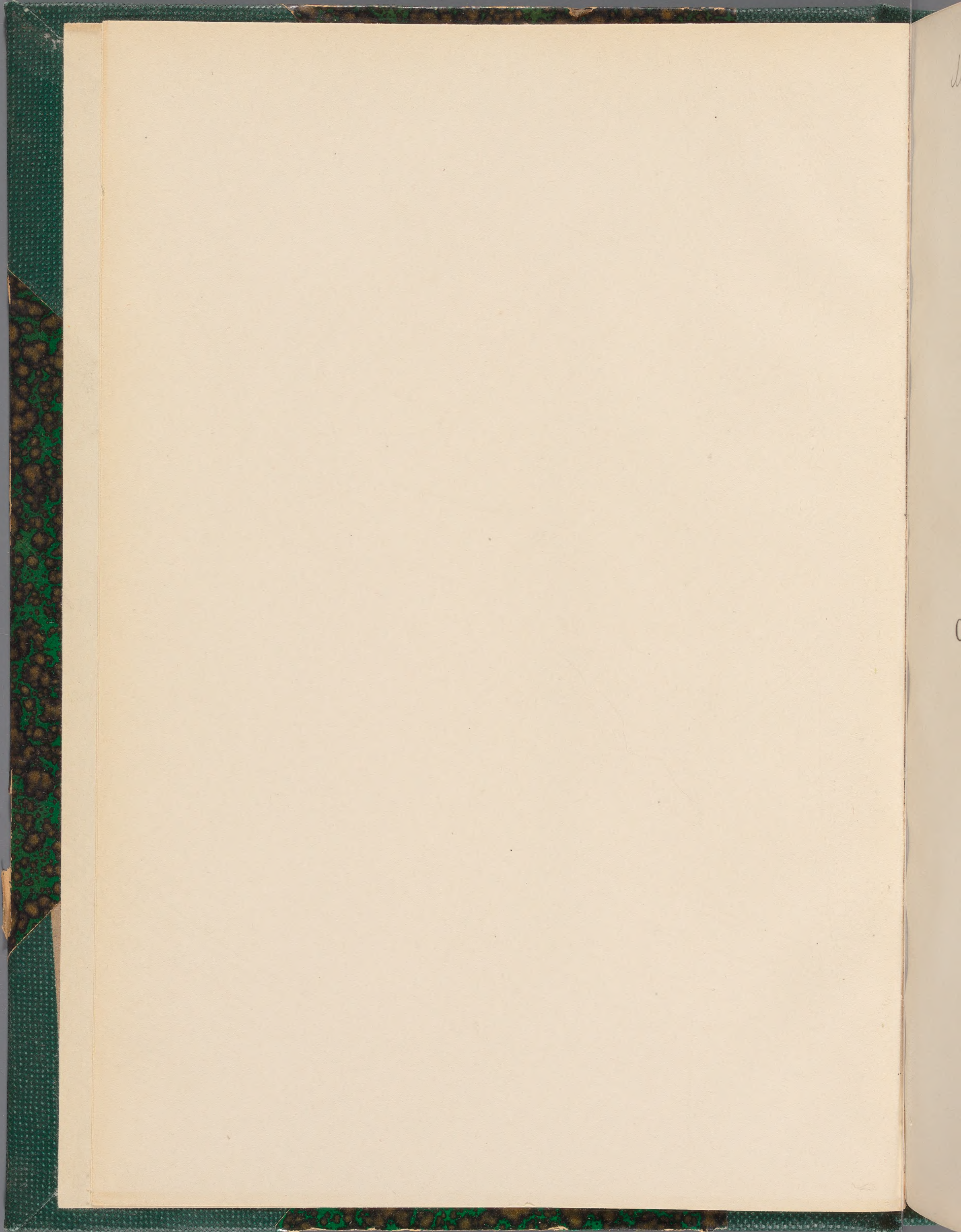
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April 4, 1910 - March 6, 1912.









Mar. 6. 1912

東京帝國大學
農科大學紀要

第二卷

JOURNAL

(FORMERLY BULLETIN)

OF THE

COLLEGE OF AGRICULTURE,

IMPERIAL UNIVERSITY OF TOKYO.

VOLUME II.

TOKYO.

Published by the University.

1909—1911.

1080
24-8

3064 mtd

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OF THE

COLLEGE OF AGRICULTURE,

IMPERIAL UNIVERSITY OF TOKYO.

TOKYO.

Published by the University.

DECEMBER 8TH, MEIJI XLII (1909).

PUBLISHING COMMITTEE.

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All communications relating to this Journal should be addressed to the
Director of the College of Agriculture.

Studies on the Chemotactic and Other Related Reactions of the Swarm-Spores of Myxomycetes.

BY

S. Kusano.

With one Figure in the Text.

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I. Introduction.

The present article embodies a full account of the results of observations on the reactions of the swarm-spores of Myxomycetes to several chemical agents, in particular on chemotaxis. After the chief results had been already published in a preliminary note ('07), I carried out further experiments which seemed to me to be of sufficient extent to justify my general conclusions already set forth and moreover to bring out the behaviour of each agent towards the chemotactic movement of the organism in a more definite way. Besides describing the results of all these experiments, it is my purpose to give a detailed account of the experiments, by which the conclusions have been reached.

Since the theory of electric dissociation has opened a wide field for the study of the physiological effect of solutions of chemical agents, numerous papers have appeared dealing with this problem. In the plant kingdom, so far as I know, an attempt was first made by BULLER ('00) on the fern-spermatozoid to make out the relation between the ionised components in solution and chemotaxis. An exhaustive study was then made by SHIBATA ('05a, '05b, '05c, '05d) on the spermatozoids of several species of Pteridophyta, which has extended considerably our knowledge of the relation between the ionised chemical agents and chemotaxis. KNIEP'S ('06) investigation on the chemotaxis of Bacteria has also been carried out in the same direction. In the animal kingdom, GARREY ('00) has published, in the same year as BULLER, the results of his observations

on the effect of ions upon the aggregation of flagellated Infusoria. The same subject was afterwards treated by JENNINGS and MOORE conjointly ('02).

We see from these investigations that the older experiments on chemotaxis by using the solutions of a certain per cent. by weight do not always bring out conclusions strictly coincident with those of the dissociation theory, or at least they do not all give us such a precise conception about the relation between the solutions and chemotaxis as is obtained by using the molecular solution, which modern chemistry demands in such a study.

When we review from this standpoint a considerable number of papers on chemotaxis published within the last decade, it is at once clear that there remains still much to be done on the chemotactic reactions, especially of the swarm-spores of Myxomycetes. Our knowledge on the chemotaxis of the last named organism is due to STANGE'S ('90) investigation made about twenty years ago. Although he has tested some number of chemicals as causing the chemotactic movement, he did not pay attention to the ionised condition of the chemicals he tested. It may be said, therefore, that it is very desirable to study the chemotactic behaviour of the swarm-spores of Myxomycetes towards some dissociable agents.

While studying the chemotactic effects of several solutions I have occasionally observed their toxic effects. Both effects of the same agent on the same organism may throw, when taken together, very clear light upon the relation of that agent to the organism. However, the account of the toxic action to be given in the present article will not extend beyond what concerns the chief aim of my present investigation. So that it is not proposed to make thorough reference to the papers dealing with the toxic action of chemicals, and I can only mention the most important and interesting papers of KAHLENBERG and TRUE ('96), HEALD ('96), STEVENS ('98), TRUE and HUNKEL ('98), and CLARK ('99). These authors have studied systematically the toxic action of chemicals upon lower and higher plants and the relative toxic value of each ion on the

different organisms studied, and have come to the conclusion that the toxicity of chemicals is due in many cases to ions and in some cases to undissociated molecules.

When free-swimming organisms react to chemicals, they show characteristic movements. This was noticed by GARREY ('00), but more extensive observations were made by JENNINGS ('97 et esq.) in lower animals and by ROTHERT ('01) in lower plants. As no particular study has hitherto been made in this respect on the swarm-spores of Myxomycetes, it seems advisable to make clear their reactions to chemical stimuli.

II. Material and Methods.

The ripe spores, brought from the field to the laboratory, were sown from time to time on distilled or tap-water in a small vessel. Numerous swarm-spores were liberated generally in a few hours and were immediately used for experiments. Of the several species of Myxomycetes collected, germination in water took place easily only in *Aethalium septicum*, *Stemonitis fusca*, and *Comatricha longa*, so that the experiments were carried out entirely with these three species. Of these, again, *Aethalium* furnished the most active spores with pronounced reactions to stimuli. Among the species which did not germinate there were those which JAHN ('05) and CONSTANTINEANU ('06) used and easily succeeded in liberating the swarm-spores in water. I did not proceed further on this point, since it was not the aim of the present study.

In the chemotactic experiment a drop of the culture-medium was brought with a pipette on the slide, and to protect the drop, and consequently the spores themselves, from mechanical disturbance a cover-glass was put over it, small pieces of glass or well-washed quartz sand being placed under it in order to leave a thick layer of water between it and the slide. The use of a cover-glass may bring about lack of oxygen in the culture-medium, but it did not show any effect on the movement and activity of the spores during a few hours' observations,

although STANGE noted in his experiments the starvation of the spores from oxygen.

In testing the action of chemicals upon the spores I followed mainly the usual capillary method of PFEFFER ('84). Capillary glass tubes about 1.5-2 cm. in length and 0.05-0.25 mm. (mostly 0.1 mm.) in calibre were filled entirely with the solution to be tested, and after sealing one end with a mixture of wax and vaseline, the other open end, previously washed quickly with water, was inserted in the culture-medium under the cover-glass. In order to prevent a violent expansion or contraction of the contents of the tube under a slight variation of temperature, care must be taken to exclude all air-bubbles from the tube, since owing to the greater coefficient of expansion of the air, they will seriously interfere with the uniform diffusion of the solution from the mouth of the tube. Indeed, as the spores are not very sensitive to oxygen, we found it needless to leave air-bubbles inside the tube, though their presense is necessary in some cases (PFEFFER, '84). It may be also noted that the capillary tube thus prepared does not show any defect and gives precisely the same results as obtained by the usually prepared tube, that is, the tube injected with a given solution by the action of an air-pump, after closing one end in a gas flame. It must be remembered that the sealing substance I used, if rather hard, does not violate in any way the chemical action of the solution in the tube. Again, in the present method very quick washing of the open end of the tube is sufficient to free it from the solution that has overflowed, while in the injection-method much more time is required to wash off the outer surface of the tube which has been spoiled by immersion in the solution. Consequently, by the former method the solution just at the mouth of the tube is kept approximately at the given strength, though by the latter it would be much diluted during washing.

Stock solutions of acids and alkalies made up on the basis of gram-molecules per litre, from which several required dilutions were made, were tested by the titration method to ascertain whether their acidity and alkalinity are accurately in the required strength.

STANGE noted that the movement of the swarm-spores of Myxomycetes was slower than that of the other free-swimming organisms used for chemotactic experiments, so that the effect of the chemotaxis appeared after a considerable lapse of time. However, in my experiments it was found that much shorter time than expected was sufficient to bring forth a pronounced effect, viz., 5-10, or more safely 20 minutes. I must also remark that under certain circumstances it is not safe to draw conclusions from observations after one hour or more, as STANGE thinks necessary; for, the diffusion-zones round the mouth of the tube become highly diluted after such a long interval of time, and without special precaution a correct notion can hardly be obtained.

In determining whether certain chemicals are attractive, repulsive, or indifferent to the spores concentrated solutions are preferably used. With a comparatively weak solution, though supraoptimal, positive results may easily be overlooked. This is due, in my opinion, to a great dilution at the diffusion-zones and to the rapid decrease of strength in these zones, by which the diffusion-zone near the mouth of the tube falls far below the critical strength for exerting attraction before the approach of the spores during their random movement. When, however, rather concentrated solutions are used, the diffusion-zone near the mouth is kept in sufficient strength for a long time, so that positive results are easily obtained.

It is well known that temperature is an important factor in determining the response of an organism to chemical stimuli. Full accounts of an example on this point was given by BROOK ('06) and ZEHL ('08) in their studies on the toxic effect of chemical agents on the germination and growth of certain fungi, showing that the same caution is requisite in my experiments, especially when the question was about the relative value of several chemicals in their action upon the spores. So that when necessary the experiments were made under a constant temperature. Further, it was necessary to carry the experiments under the optimal temperature for the activity or sensibility of the spores, which was found to be 20-25°C. For these purposes the observations were made in the

majority of cases in a greenhouse where the temperature was nearly constant ($20^{\circ}\text{C}.$) during the experiments.

In the present study the diameter of the capillary tube causes very divergent effects upon the collection of the spore near its mouth, and for the comparative study of different chemicals or of different concentrations of a chemical it is necessary to use tubes of approximately equal diameter.

Unless otherwise stated, the experiments were made with the materials of the same collection. The different collections of the same species may behave very divergently towards the same chemical stimulus.

PFEFFER ('88, p. 584) has noted several important precautions necessary for chemotactic experiments by the capillary method, to all of which due attention has been paid during the present experiments.

III. General Physiology of the Swarm-Spores.

Before entering into the proper subject of the present investigation it will be necessary to give a brief account of some physiological phenomena of the swarm-spores bearing more or less upon our present study.

1. GERMINATION OF THE SPORES.

Germination takes place as well one year after collection as when just mature. Sown on water the dried spores are hardly moistened, and all float on the surface. If the air in contact with the water be kept dry, germination is delayed considerably. If, however, the air be kept sufficiently moist, it is greatly accelerated. Thus under the latter condition, *Aethalium* germinates at $20^{\circ}\text{C}.$ in 2.5 hours, while under the former condition, it remains unchanged even after 20 hours at the same temperature. *Stemonitis* and *Comatricha* behave also quite similarly. If the floating spores are brought under the cover-glass or in a moist chamber, they begin to germinate immediately. It follows that the moist air

promotes germination. It seems to me that the moist air facilitates the absorption of water by the spores and the rapid absorption of water brings about the rapid germination.

Generally, a high temperature promotes germination, but it is not entirely prevented at a low temperature. In a sufficiently moist chamber I was able to raise the swarm-spores of *Aethalium* on water at 1-10°C.

In my experiments the time needed for germination has varied with different gatherings of the same species, as also observed by LISTER ('91).

It is a very striking fact that acidic substances acts favourably on germination. If, for instance, a capillary tube containing malic acid of 1 mol be inserted in the water under the cover-glass in which ungerminated spores of *Aethalium* are kept uniformly scattered, it will be observed that the spores at certain diffusion-zones of the acid begin to germinate earlier than others. Thus one hour after insertion, the spores at 5 mm. distance from the mouth of the given tube has began to germinate, while those beyond did not. This was the most frequent occurrence during my chemotactic experiments with several other acids. This appeared so remarkable that special experiments were carried out to ascertain the fact more accurately. They are given below:

Experiment 1 (March).

Aethalium: sown in open glass vessels, exposed to direct sunlight.

Culture-Medium.	Observed after one hour.	Observed after 2 hours.
Sulphuric acid 1/1200 mol.	Medium cloudy with swarm-spores.	Almost all germinated.
Acetic acid 1/1000 mol.	Medium clear; no germination.	Partly germinated.
Tap-water.	Medium clear; no germination.	No germination.

Remark:—In acidic fluids complete moistening of the spores is effected sooner than in tap-water.

Experiment 2 (March).

Aethalium: prepared as before.

Culture-medium.	Observed after 30 minutes.	Observed after 1½ hours.	Observed after 2 hours.
Malic acid 1/800 mol.	Spores completely moistened and suspended in the medium; no germination.	Spores begin to germinate.	Medium cloudy with swarm-spores.
Malic acid 1/1600 mol.	Spores partly moistened and suspended in the medium; no germination.	No germination.	Medium less cloudy.
Tap-water.	Spores quite unmoistened and all float on the surface of the medium; no germination.	No germination.	Few spores germinated; the other begin to germinate.
Tap - water. (spores moistened previously with alcohol).	Spores immediately suspended in the medium; no germination.	Spores begin to germinate.	Medium cloudy with swarm-spores.

Experiment 3 (May).

Aethalium one year old, and *Stemonitis* two years old. Vessels were kept in a sufficiently moist air under a bell-jar. Temperature 25-28°C.

Culture-medium.	<i>Aethalium</i> , observed after one hour.	<i>Stemonitis</i> , observed after one hour.
Hydrochloric acid 1/1000 mol.	Nearly half of the spores germinated; medium cloudy.	Spores moistened, but no germination.
„ 1/5000 „	Spores partly germinated, not yet wholly moistened.	Spores moistened, but no germination.
„ 1/10000 „	Germination less than in 1/5000 mol; not moistened.	Spores moistened, but no germination.
„ 1/50000 „	Germination very few; not moistened.	Spores moistened, but no germination.
Distilled water.	No germination, not moistened.	Spores moistened, but no germination.

Remark:—*Aethalium* began to germinate in distilled water after 1 hour and 45 minutes, while in hydrochloric acid germination took place sooner; *Stemonitis* did not germinate after 3 hours, but the next morning swarm-spores were observed in all the vessels.

Experiment 4 (June).

Aethalium: one year old; temperature 19-20°C.; vessels open.

Culture-medium.	Observed after one hour.	Observed after two hours.	Observed in the next day.
Hydrochloric acid 1/1000 mol.	Spores began to germinate; moistened.	Medium cloudy as in distilled water.	All swarm-spores active.
Hydrochloric acid 1/700 mol.	Spores about to burst forth; moistened.	Nearly same as in 1/1000 mol.	All swarm-spores active.
Hydrochloric acid 1/500 mol.	No germination; moistened.	Very few spores germinated.	Only few spores germinated.
Sodium hydroxide 1/5000 mol.	Spores about to burst forth; not moistened.	Spores mostly germinated, but less than in 1/1000 HCl.	Swarm-spores active.
Sodium hydroxide 1/1000 mol.	Spores began to germinate, but not moistened.	Swarm-spores as much as in 1/1000 HCl., but less active.	Many swarm-spores active.
Oxalic acid 1/1000 mol.	Spores began to germinate; moistened.	As in 1/1000 HCl.	Many swarm-spores active.
Tap-water.	Very few spores germinated; not moistened.	Few spores germinated.	Many spores germinated.
Distilled water.	Very few spores germinated; not moistened.	Many spores germinated, and active.	Swarm-spores mostly active.

Remark:—Hydrochloric acid 1/700-1/1000 mol and oxalic acid 1/1000 mol are the critical strengths for germination.

Experiment 5 (June).

Aethalium: one year old; sown on drops on slides under a bell-jar;
temperature 20-22°C.

Culture-medium.	Observed after 35 minutes.	Observed after 1 hour and 20 minutes.	Observed after 2 hours.
Malic acid 1/1000 mol.	Germination begins.	More than half of spores bursted forth.	Swarm-spores active.
Malic acid 1/500 mol.	„	More than half of spores bursted forth, not yet active.	„
Tartaric acid 1/1000 mol.	No germination.	Many spores germinated, but not so much as in malic acid.	?
Tartaric acid 1/500 mol.	„	Very few spores germinated.	?
Acetic acid 1/1000 mol.	Spores begin to burst forth.	Many spores germinated, but mostly still inactive.	Swarm-spores become active.
Acetic acid 1/500 mol.	Many spores bursted forth.	More than half of spores bursted forth, but mostly still inactive.	Swarm-spores active.
Boric acid 1/10 mol.	No germination.	No germination.	No germination.
Boric acid 1/5 mol.	„	„	„
Tap-water.	Very few spores began to germinate.	Many spores germinated.	—
Distilled water.	No germination.	More than half of spores germinated.	Swarm-spores active.

Experiment 6 (June).

Aethalium: one year old; sown on slides; temperature 20-22°C.

Culture-medium.	Observed after 45 minutes.	Observed after 3 hours and 30 minutes.
Malic acid 1/1000 mol.	Many spores germinated and swarm-spores began to swim about.	Medium cloudy with swarm-spores.
Boric acid 1/20-1/30 mol.	Very few spores germinated.	Many spores germinated.
Ammonia 1/1000 mol.	No germination.	No germination.
Ammonia 1/500 mol.	No germination.	No germination.
Distilled water.	Many spores germinated and swarm-spores began to swim about.	Medium cloudy with swarm-spores.

These experiments lead to the conclusion that the spores germinate somewhat sooner in acidic solutions than in water. This seems to be due to the moistening action of the acids, stronger solutions moistening more rapidly than weaker ones. Moistened previously with weak alcohol the spores germinate just as soon as in acidic solutions. This shows clearly the connection of germination to the moistening of the spores.

CONSTANTINEANU ('06, p. 502) observed an unfavourable action of acids upon the germination of many Myxomycetous spores. This contrary result is probably due, in my opinion, to the strong solutions he used. For, I could also observe in my experiments the same action of concentrated acids.

The action of the acids just mentioned was more evident when the experiments were carried out at a comparatively low temperature.¹ At a high temperature, say 20-25°C., germination took place in tap-

1. For the relation of temperature to the germination of the spore refer to JAHN'S ('05) paper.

water and distilled water nearly as soon as in acid solutions, the time difference being reduced almost to *nil*. Even at low temperatures similar results were obtained, if the surrounding air was kept sufficiently moist.

The promotion of germination by acids has not been conclusively shown for *Stemonitis* and *Comatricha*, for which a further research is necessary. I may however mention that in *Stemonitis* germination is promoted by sodium chloride, which shows no effect on other Myxomycetes. At the right strength of this salt the spores of *Stemonitis* are moistened easily and liberate the swarm-spores earlier than in water, just as *Aethalium* does in acids.

2. TEMPERATURE AND ACTIVITY OF THE SWARM-SPORES.

During my experiments extending over several months I became struck with the fact that the activity of the swarm-spores as well as their reaction to chemical stimuli varied much according to different hours of the day and different days of the month. Thus the place of observation remaining the same, the swarm-spores were generally less active in the forenoon, especially in the earlier hours, and most active in the afternoon till about 3 p.m. (September-October). Their activity and sensitiveness diminished with the approach of evening.

Similar variation was also observed in different weathers. It was exhibited most remarkably in midsummer (August): on a moist hot day (max. temp. 30°C. in the room) the spores reacted most sensitively, while on a cool rainy day of the same season the reaction was markedly feeble.

In their experiments of chemotaxis VOEGLER ('91, p. 673) and STANGE ('90, p. 139) have found a similar relation between temperature and the sensibility of the organisms. This has been ascertained by BROOK ('06) in other response of organisms to chemical agents. In my experiments the slow movement may affect on the chemotactic response to some extent, yet the diminished sensitiveness at a low temperature must

be looked upon as the chief cause of feeble attraction or repulsion.¹

Repeated experiments have shown that 20-25°C. is the optimum temperature for the sensitiveness of the spores. At a temperature over 30°C. their body may shrink and become immovable. Again the movement becomes slow and the sensitiveness lessened at a temperature of 10-12°C. The influence of temperature on the spores was easily observed directly under the microscope, by cooling the culture-medium with a piece of ice. When the medium was cooled below 5°C. the spores became immovable with shrunken body, but recovering soon when the temperature was raised. The range of temperature within which an approximately similar effect could be obtained was 20-25°C. Hence experiments on chemotactic and other reactions of the swarm-spores must be made within this range of temperature.

3. OXYGEN AND ACTIVITY OF THE SWARM-SPORES.

As already remarked by STANGE, the influence of oxygen on the chemotactic activity of the swarm-spores is not apparent. Under the cover-glass they swim for a few hours as actively as in an open culture-medium. Hence it appears that no precaution is necessary to keep the medium thoroughly aerated during the chemotactic experiment, so that I have carried it out under the cover-glass. I can easily cite several proofs of comparative indifference to oxygen: the spores remained as active in a capillary tube containing no air-bubble, for one hour or more at 2-5 mm. distance from the mouth of the tube, as they were outside the latter; under the cover-glass they were indifferent to the air-bubbles present in the medium; spores densely sown in water under the cover-glass could germinate in a few hours or next day, apparently irrespective of the absence or presence of oxygen; the germination took place in closed vessels or in boiled water equally well as in open vessels or in fresh tap-water; the

1. The assertion that the variation of the response in this case is strictly connected with the changes of temperature was justified by the fact that the effect was always constant under constant temperature, irrespective of the intensity of light which may vary widely at different hours of the day and in different weathers.

swarm-spores thus hatched out were all active and could be employed for experiment, etc. (cf. Zopf, '85).

4. LENGTH OF THE SWARM-PERIOD.

In his study of Myxomycetes LISTER ('91) has mentioned that the swarm-spores become myxoamoebæ in a few days and small plasmodium afterwards. On the whole, the swarm-period is here considered to be longer than that of any spermatozoid (PFEFFER, '04, p. 703). In my experiments the swarm-spores continued active for a few days without becoming myxoamoebæ. As far as observed by me, the swarm-spores of *Aethalium* may remain alive in tap-water for two weeks; those of *Stemonitis* and *Comatricha* may also swarm for nearly the same length of time. These results have been obtained by the following experiments:

The vessels containing the swarm-spores were placed at different places in different temperatures during the winter months, and the conditions of the spores were noticed day after day. The spores were all derived from the same collection.

I. Greenhouse. (temperature 18-21°C.).

a. *Aethalium*. Many swarm-spores having perished, there remained only a few in active state after 5 days. The culture-medium was spoiled by Infusoria. It seemed likely that the number of surviving spores depended upon the number of the Infusoria developed: while the latter was comparatively few in number, the swarm-spores were found mostly active. This shows that Infusoria devour the Myxomycetes. After 10 days there were no spores.

b. *Stemonitis*. After 5 days there were numerous Bacteria in the culture-medium, but no Infusoria was observed. The swarm-spores were almost all active. After 10 days, however, there were no living spores.

c. *Comatricha*. After 5 days Infusoria and Bacteria spoiled the culture-medium, so that many of the swarm-spores have perished. After 10 days a few spores were found alive.

II. Laboratory room. At noon the temperature rose to 20-22°C., while at night it sank to nearly 1°C. The swarm-spores swam about

during the day-time, but sank to the bottom of the vessel at night, their body becoming shrunken. When the temperature again rose the next day, the body again became plump and the swimming state was resumed.

a. *Aethalium*. The number of the active spores was not remarkably reduced after 12 days.

b. *Stemonitis*. Almost all were active after 12 days.

c. *Comatricha*. Some spores were active after 12 days.

III. Cold room. The temperature fluctuated between 9 and 1°C., and one morning ice-formation was observed on the surface of the culture-medium. At the lower temperature the spores rounded themselves up and became motionless. If such spores were brought into the room at 20°C. they began to move first like an amoeba and then recovered the usual form of the swarm-spore. During the experiment I was not able to find encysting, and the shrunken form represented merely the resting condition of the swarm-spores.

a. *Aethalium*. Most of the spores were active after 12 days as before, if they were often brought into the room at 20°C.

b. *Stemonitis*. Most of them perished after 12 days. The surviving ones were, however, very active.

c. *Comatricha*. Very few spores lived for 12 days after hatching.

It will be seen from the above experiment that the swarm-spores can be active for about two weeks without metamorphosing into myxoamaeba or microcyst at a comparatively low temperature. At a higher temperature some microorganisms spoil the culture-medium, and their presence seems to be disadvantageous to the existence of the swarm-spores of Myxomycetes.

5. RELATION TO OTHER ORGANISMS.

In natural condition Myxomycetes feed mostly on rotten wood or leaves, which may also furnish the food-materials to Bacteria, yeast, Infusoria, etc., and it is easy to see that there must occur a struggle for existence among these organisms. It is, therefore, biologically interesting to see in what relation Myxomycetes stand to other organisms in this

respect. LISTER ('90, '91) has already reported that the swarm-spores of Myxomycetes digest Bacteria (cf. PINOY, '02, '07). MILLER ('99, p. 60) found that the swarm-spores multiply at the expense of Bacteria. CHIRZASZCZ ('02) observed that the swarm-spores digest yeast. It follows that the existence of such organisms in the culture-medium of Myxomycetes would favour the development of the latter. In accordance with this view I have found, as already stated, that Bacteria developed in the culture-medium did not prevent in any way the swimming of the swarm-spores. Moreover, I observed that, when a certain bacterial colony was added to the culture-medium, the swarm-spores appeared to become more active than before. This seems to be due probably to their consuming the Bacteria. Thus, while the presence of many organisms has rather favourable effect on the swarm-spores, certain Infusoria, as far as observed in my experiments, may have an unfavourable effect. It is quite certain that Infusoria devour the active swarm-spores (LISTER, '01): as soon as the culture-medium becomes contaminated with this animal, the swimming swarm-spores clearly diminish in number. This is proved by the fact that on addition of the Infusoria to the culture-medium, the cloudy condition due to the presence of immense number of the spores disappears, and the medium becomes transparent in a day. The rapid decrease of the active swarm-spores in the medium kept at a higher temperature, as shown in the foregoing experiment, is due probably to the development of the Infusoria and their destructive action.

IV. Chemotaxis.

1. FREE ACIDS.

STANGE has already observed that the swarm-spores of Myxomycetes are attracted by acids. His studies were made chiefly on *Chondrioderma difforme* and *Aethalium septicum*, in which he found that the attractive action was confined to certain kinds of organic acids and that their action

was somewhat different according to the different species of Myxomycetes. Thus he found that *Chondrioderma* was attracted by malic acid more remarkably than by lactic and butyric acids, while *Aethalium* was more sensitive to lactic, butyric, valerianic, and propionic than to malic acid. What is more remarkable, while he ascertained that some neutral salts of the acids mentioned above had likewise attractive action, he could not succeed in finding out any inorganic acid or other organic acids that attracted the spores. Hence his results would lead us to the conclusion that the chemotaxis is connected with the constitution of these acids, undoubtedly with anions according to the dissociation theory. This is a clear and definite conclusion from his experiments, which needs no further explanation. Following in STANGE's line, I undertook first of all to test the chemotactic action of as many kinds of acids as I could get access to. The experiments were made with *Aethalium*, *Stemonitis*, and *Comatricha* in preparations similar to those of STANGE. It must be especially remarked that I have here chosen the same species of *Aethalium* as STANGE used, if my identification is correct.¹ At first sight it appeared as if all the organic acids which were found by STANGE to be stimulant exerted attraction on all the species of Myxomycetes I used. Repeated experiments, however, have shown that there was no such restricted relation between the organism and acids as STANGE supposed. Indeed, all the organic and inorganic acids I tested had a similar effect upon the spores, viz. positive chemotaxis. In moderate concentrations the spores reacted rapidly to acids and their collection near the mouth of the capillary tube was clearly observed in 5-10 minutes. In the case of more concentrated solutions collection near the mouth was not observed in 1-2 hours, but afterwards the dense collection became evident. Of the three species of Myxomycetes *Aethalium* reacted most rapidly, so that the effect of the acids became evident in 30 minutes, while with the less sensitive and perhaps less active *Stemonitis* 1-2 hours elapsed often before the result became apparent. *Comatricha*, which has the largest swarm-spores of the

1. For identification I depended on LISTER's "Mycetozoa. 1894."

three species, seems less active in movement than *Stemonitis*, but the reaction was more rapid than in the latter.

As has been already noticed, it is advisable to use a rather concentrated solution of acids in order to obtain indubitable results. For this reason I used generally 1/5-1/20 mol. The list of acids tested and their relative effects in equimolecular concentrations upon the three species of Myxomycetes are given in the following table. Here a marked attraction is denoted by A, a weak attraction by a, and an ambiguous effect by ?, while — shows that no experiment was made.

Table I.

Acid.	Reaction of swarm-spores.		
	<i>Aethalium.</i>	<i>Stemonitis.</i>	<i>Comatricha.</i>
Hydrochloric acid HCl	A	A	A
Nitric acid HNO ₃	A	A	A
Sulphuric acid H ₂ SO ₄	A	A	A
Phosphoric acid H ₃ PO ₄	A	A	A
Chromic acid H ₂ CrO ₄	A	A	A
Boric acid H ₃ BO ₃	a	a	?
Formic acid H.CO ₂ H	A	A	A
Hydrocyanic acid HCN	a-?	a-?	—
Acetic acid CH ₃ .CO ₂ H	a-A	a-A	a-A
Propionic acid CH ₃ CH ₂ .CO ₂ H ...	a-A	a-A	a-A
Butyric acid CH ₃ (CH ₂) ₂ .CO ₂ H...	a-A	a-A	a-A
Valerianic acid CH ₃ (CH ₂) ₃ .CO ₂ H...	a-A	a-A	a-A
Lactic acid C ₂ H ₄ .OH.CO ₂ H ...	A	A	A
Oxalic acid (CO ₂ H) ₂	A	A	A
Succinic acid C ₂ H ₄ .(CO ₂ H) ₂ ...	A	A	A
Malic acid C ₂ H ₃ .(OH).(CO ₂ H) ₂	A	A	A
Tartaric acid C ₂ H ₂ .(OH) ₂ .(CO ₂ H) ₂	A	A	A
Citric acid C ₃ H ₄ .OH.(CO ₂ H) ₃	A	A	A
Picric acid C ₆ H ₂ (NO ₂) ₃ (OH) ...	A	?	?
Salicylic acid C ₆ H ₄ (OH).CO ₂ H ...	A	?	a
Tannic acid	a	?	?

From this table it will be seen that the attraction by acids is a universal phenomenon for the swarm-spores of Myxomycetes.

Now, to show the manner of reaction of spores to the different concentrations of acids an example may be given as follows:

The swarm-spores of *Aethalium* was exposed to the action of malic acid contained in the capillary tube of 0.1-0.13 mm. in diameter, and their reaction was observed after 10-20 minutes.

1/2-1/5 mol.—Cloudy collection of the spores at a certain distance from the mouth of the tube.

1/15-1/20 mol.—Immense cloud around the mouth, but none in the tube.

1/50 mol.—The spores filled up the mouth, and densely crowded round it.

1/100 mol.—Collection round the mouth less dense; the spores filled up the mouth and many of them entered the tube as far as 0.5 mm. from the mouth.

1/200-1/300 mol.—Diffuse cloud in the mouth, filling the tube 0.3-0.6 mm. deep.

1/400 mol.—Collection round the mouth not clear; few spores entered the tube.

1/500 mol.—Indifferent.

In equimolecular concentrations the reaction of the spores varies according to the kind of acids. For instance, in its action 1/20 mol sulphuric acid corresponds to 1/2 mol malic acid, and 1/50-1/100 mol to 1/10-1/20 mol and 1/600 mol to 1/400 mol respectively. 1/5-1/20 mol acetic and lactic acids correspond just to the same concentrations of malic acid, but their 1/200 mol exerts a stimulus comparable to that of 1/400 mol of malic acid, while their 1/300 mol behaves quite indifferently. Again, the attraction of 1/20-1/50 mol hydrochloric acid is comparable to that of the same concentration of sulphuric acid, but 1/300 mol is quite indifferent.

Picric and salicylic acids are less soluble in water, and their concentrated solutions could not be tested. Their saturated solutions (picric

acid *ca.* 1/4, salicylic acid *ca.* 1/50 mol) attracted apparently the spores round and inside the tube just as strongly as 1/50 mol malic acid.

Boric acid is very weak. Its saturated solution (*ca.* 2/3 mol) can hardly attract. In *Stemonitis* and *Comatricha* the reaction seemed to be uncertain, but in *Aethalium* it was very definite.

Hydrocyanic acid is weaker than boric acid, and its attraction was far more obscure.

It appears after all that the swarm-spores of Myxomycetes are positively chemotactic towards acids. The intensity of reaction accords well with the degree of acidity, so that in equimolecular concentrations the stronger acids, such as inorganic acids, act more strongly than the weaker ones, and dibasic acids more strongly than monobasic acids, provided the degree of dissociation is equal. These facts are therefore sufficient to prove that the attraction of the spores is simply concerned with the acidic character of the acids, no matter what their constitution may be.

2. ACIDIC SALTS AND ACIDIC SUBSTANCES.

Acidic salts as well as such salts as give acidic property to the aqueous solution by hydrolysis act upon the swarm-spores similarly as the free acids, all attracting the spores markedly in moderate concentrations. The salts tested were the following:

Acid calcium malate.

Primary calcium phosphate.

Monopotassium phosphate.

Potassium bisulphate.

Sodium bisulphate.

Potassium bichromate.

Aniline sulphate.

Aniline chloride.

The manner of attraction of the spores by these chemicals differs somewhat from that of the free acids mentioned above. With free acids in concentrated solutions no spores were observed to enter the tube after assembling around its mouth. However, in equimolecular concentrations

the salts attract the spores into the tube, so that they swim about diffusely in it. A similar difference between certain acids and their salts as regards the attraction of the swarm-spores of *Chondrioderma* was observed by STANGE (p. 158). The salts he tested were however all neutral, so that the respective relation of the acids and the salts to the spores must be different in this case.

In connection with this fact we may mention that the attracting power of the salts is not much feeblener than that of free acids. For example, 1/10 mol malic acid and its acid calcium salt attract nearly equal number of the spores of *Aethalium*, when the tubes containing these chemicals are inserted into the same culture-medium: after 30 minutes the same dense cloud of spores round and in the mouth may be observed, though the spores advance deeper into the tube containing the salt. To show this relation more clearly I will note down some experiments made with *Aethalium*.

Experiment 1.

Diameter of the tube 0.1-0.15 mm.; cool and rainy day in July.

1.—Observed after 10 minutes.

Phosphoric acid: 1/10-1/15 mol.—Dense cloud of the spores round the mouth, leaving the centre empty.

1/20-1/30 mol.—Small collection round the mouth.

Monopotassium phosphate: 1/10 mol.—Remarkable collection at the mouth, and deep and dense entry into the tube.

1/20 mol.—The same.

Tripotassium phosphate: 1/10 mol.—Apparent repulsion.

1/20 mol.—Slight repulsion.

2.—Observed after 40 minutes.

Phosphoric acid: 1/15-1/30 mol.—No entry into the tube.

Monopotassium phosphate: 1/10-1/20 mol.—Cloudy entry 5 mm. deep into the tube, motion active.

3.—Observed after 40 minutes.

Phosphoric acid: 1/15 mol.—Cloudy collection at and round the

mouth, but no entry into the tube.

Monopotassium phosphate: $1/5$ mol.—Cloudy collection at the mouth, and the mouth nearly filled up by the spores.

4.—Observed after 10 minutes.

Phosphoric acid: $1/2$ mol.—The spores form a ring round the mouth 2 mm. in diameter.

Monopotassium phosphate: $1/2$ mol.—Large cloud round the mouth; no repulsion-space at the mouth and no entry into the tube.

Tripotassium phosphate: $1/2$ mol.—Remarkable repulsion.

5.—Observed after 10 minutes.

Phosphoric acid: $1/70$ mol.—Somewhat diffuse collection; no entry into the tube.

Monopotassium phosphate: $1/50$ mol.—Small but dense cloud at the mouth and gradual entry.

$1/60$ mol.—Weak attraction.

Experiment 2.

Diameter of the tube 0.1-0.13 mm.; warm day in October, temperature being somewhat lower than in the former experiment.

1.—Observed after 20 minutes.

Phosphoric acid: $1/70$ mol.—Dense cloud round the mouth; few spores enter the tube, coming to rest sooner or later.

Monopotassium phosphate: $1/50$ mol.—Cloud round the mouth but less dense. Numerous spores enter the tube; the spores in the tube turn back at the mouth, and advance inwards into the tube.

Tripotassium phosphate: $1/20$ mol.—Indifferent or slight repulsion.

Experiment 3.

Diameter of the tube 0.1-0.13 mm.; the same day as Experiment 2.

1.—Observed after 4 minutes.

Chromic acid: $1/5$ mol.—Repulsion-space in the centre of the cloud round the mouth.

Potassium bichromate: $1/5$ mol.—Immediate collection round the mouth; no entry.

Potassium chromate: $1/5$ mol.—Indifferent or repulsion.

The same tubes observed after 20 minutes.

Chromic acid: $1/5$ mol.—The spores approached the mouth; yet a small repulsion-space was found in front of the mouth.

Potassium bichromate: $1/5$ mol.—Numerous spores entered the tube.

Potassium chromate: $1/5$ mol.—Indifferent.

2.—Observed after 5 minutes.

Chromic acid: $1/50$ mol.—Cloudy collection at the mouth.

Potassium bichromate: $1/50$ mol.—Cloudy collection at the mouth.

After 10 minutes.

Chromic acid: No entry at all.

Potassium bichromate: Entry of immense number of spores.

3.—Observed after 6 minutes.

Chromic acid: $1/100$ mol.—Collection at the mouth remarkable.

Potassium bichromate: $1/100$ mol.—Collection less remarkable than in the acid.

4.—Observed after 7 minutes.

Chromic acid: $1/150$ mol.—No collection at the mouth, but cloud of spores in the tube near its mouth.

Potassium bichromate: $1/150$ mol.—No collection at the mouth, but the spores enter the tube freely.

In the case of acids the spores entering the tube soon stop after reaching to a certain distance from the mouth and reverse the direction of their movement, swimming about to and fro within a certain section of the tube. All the spores are thus gathered in a limited portion of the

tube forming a cloud. However, in the case of salts the spores proceed deeper and deeper unarrested in their movement, and they reverse the direction of their motion the moment they come out of the tube. After 30 minutes they are distributed in the tube within the length of 4-5 mm.

5.—Observed after 30 minutes.

Chromic acid: 1/300 mol.—No attraction (that the spores react to this concentration at a higher temperature will be seen in later experiments).

Potassium bichromate: 1/300 mol.—No attraction.

It is evident from these experiments that in a dilute solution the acid and its salt have nearly a similar action upon the spores, but in concentrated solutions the acids acts repulsively to a degree that its salt does not. This shows that in high concentrations the acid contains a comparatively larger amount of a repellent component or components than its salt.

The same behaviour of acids and their salts as observed in *Aethalium* was found also in *Stemonitis* and *Comatricha*.

Besides the acidic solutions of chemicals I tested the action of acidic substances taken from the plant tissue. The juices of apple, grape, *Citrus*, and *Punica* fruits, and of the stem and leaves of *Rumex* are powerful agents of the positive chemotaxis. It is interesting that boiling water extract of decayed wood has the same action upon the spores. The extract gave acidic reaction, so that the acidity must be concerned in this case.

So far experiments with several acidic substances confirm my conclusion drawn from the results of the experiments with free acids that the positive chemotaxis is connected with the acidity of the agents.

3. NEUTRAL SALTS AND NEUTRAL SUBSTANCES.

Inorganic as well as organic salts of alkali metals, alkali earths, and the magnesium group were tested in the same concentration as the free acids and other acidic substances. None of them had an attracting action upon the spores of *Aethalium* and *Comatricha*. What is remark-

able, however, is that *Stemonitis* reacted to calcium chloride, sodium chloride, potassium sulphate, and zinc sulphate, and was attracted by them, while it was quite indifferent towards other salts. The attracting action here was not so remarkable as in the free acids or acidic substances, and it is a question in my mind whether we have before us actual chemotaxis, or only an apparent collection due to other causes pointed out by PFEFFER ('04, p. 754).

Generally speaking, neutral salts, unless they are not highly poisonous, do not call forth in moderate concentrations any chemotactic movement of the swarm-spores. The salts tested are given in the following table:

Table II.¹

Salt.	<i>Aethalium.</i>	<i>Stemonitis.</i>	<i>Comatricha.</i>
Sodium chloride	×	Attraction.	×
Potassium chloride	×	—	—
Ammonium chloride	—	×	—
Barium chloride	×	×	—
Calcium chloride	×	Attraction.	×
Potassium nitrate	×	×	—
Barium nitrate	×	—	—
Sodium sulphate	×	—	—
Potassium sulphate	×	Attraction.	—
Zinc sulphate	×	Attraction.	—
Magnesium sulphate... ..	—	×	—
Sodium phosphate	×	×	×
Sodium acetate	×	—	—
Potassium oxalate	×	—	×
Sodium tartarate	×	—	—
Ammonium tartarate	×	—	—
Sodium potassium tartarate	×	—	—

Heavy metallic salts, such as copper sulphate and mercuric chloride,

1. Tested cases are marked with ×.

which are known as highly toxic upon organisms, are strongly repellent. So also chloroform and chloral hydrate.

Certain organic substances, such as glycerin, urea, cane-sugar, fruit-sugar, milk-sugar, ethyl alcohol, and pepton, were tested without any positive result.

4. 'ALKALINE' AGENTS.

The agents which give alkaline reaction to their aqueous solutions were tested in various strengths. They all repelled the spores, the intensity of the repulsion corresponding to the intensity of alkalinity. These agents are the following:—Sodium hydroxide, potassium hydroxide, ammonium hydroxide, potassium bicarbonate, dipotassium phosphate, tripotassium phosphate, potassium chromate, etc.

5. DISCUSSION OF RESULTS.

When we look over the results of the experiments given in the foregoing pages, the following general conclusions appear justified: (1) acidic solutions exercise positive chemotaxis, (2) alkaline solutions exercise negative chemotaxis, and (3) solutions of neutral substances, whether dissociable or undissociable, have no chemotactic action on the swarm-spores of the Myxomycetes thus far examined by me. These conclusions are in strict accordance with that given by JENNINGS ('97) in his studies on the reactions to chemicals of ciliated Infusoria. We may now inquire for the component of solutions that is concerned in the reaction. I shall in this place endeavour to solve this important question on the basis of the theory of electric dissociation.

Before going further let us pause a while on the relation between acidic substances and free-swimming organisms as brought out by previous investigations. Positive chemotaxis towards acids is known in many cases:

Spermatozoid of ferns by malic and maleic acids (PFEFFER, '84, VOEGLER, '91, BULLER, '00).

Spermatozoid of *Selaginella* by malic acid (PFEFFER, '84).

Spermatozoid of *Isoetes* by malic, succinic, fumaric, and tartaric acids (SHIBATA, '05a).

Spermatozoid of *Equisetum* by malic acid (SHIBATA, '05c, '05d).

Spermatozoid of *Lycopodium* by citric acid (BRUCHMANN, '09).

Swarm-spore of *Saprolegnia* by phosphoric acid (STANGE, '90).

Swarm-spore of *Myxomycetes* by malic, lactic, and butyric acids (*Chondrioderma*); tartaric, malic, lactic, butyric, valerianic, and propionic acids (*Aethalium*); and lactic acid (*Stemonitis*) (STANGE, '90).

Chlamydomonas by nitric acid (FRANK, '04).

Euglena by citric, lactic and phosphoric acids (FRANK, '04).

Chilomonas by acetic, butyric, and lactic acids (GARREY, '00).

Cyclidium, *Colpidium*, *Paramecium*, and *Chilomonas* by carbonic and other inorganic as well as organic acids (JENNINGS and MOORE, '02).

The attracting component of acids was not ascertained in all of these cases. The few authors who have touched upon this point have, however, expressed different views on the basis of their own observations. Thus, according to PFEFFER ('04), BULLER, SHIBATA, and BRUCHMANN, the anions of the acids are concerned in the attraction of the spermatozooids they studied. GARREY stated that *Chilomonas* is also attracted by the same component of the acids he ascertained as exerting chemotactic stimulus. This conclusion of GARREY seems to have been based on the fact that the salts of the attractive acids attract equally well as the free acids and that many others acids have no chemotactic effect. JENNINGS and MOORE ('02) investigated afterwards the reaction of the same organism to chemicals and extended also his studies to *Cyclidium*, *Colpidium*, and *Paramecium*. Their general results are that all organic as well as inorganic acids exert definitely chemotactic stimulus. The conclusion to be drawn from these results can not be other than that the cations—H-ions—of the acids are the essential factors in chemotaxis, a conclusion quite opposed to GARREY'S view. SHIBATA ('05d) has also ascertained in the spermatozoid of *Equisetum*

the attracting action of the H-ions. While it is generally admitted that the H-ions are toxic on lower and higher organisms (PFEFFER, '04, CZAPEK, '05) on the one hand, and act repellently towards most chemotactic organisms on the other (PFEFFER, '04, p. 808, SHIBATA, '05a), this fact found by JENNINGS and MOORE should be of great interest from a physiological point of view.

In his study of the swarm-spores of *Saprolegnia* STANGE has pronounced the view that the free molecules of phosphoric acid may attract them (p. 127), but we may with good reason assume that he did not pay special attention to the existence of ions in his solution; hence his conclusion can not be looked upon as valid.

The other authors have not expressed any view on the active component of the solutions, but on the basis of the results obtained by them it appears that positive chemotaxis is due mainly to the anions of the acids.

Returning to our proper subject, it must first of all be borne in mind that there may exist three components in the solutions of most of the substances I used, which are, according to the dissociation theory, anions, cations, and undissociated molecules, all having different chemical effects. To elucidate which of these components plays the principal rôle in chemotaxis, we shall institute a comparison, e.g. between hydrochloric acid, sodium chloride, and sodium hydroxide, all acting differently upon the spores, as shown above. As these are all highly dissociable agents, anions and cations must practically be the only components present in dilute solutions—H-and Cl-ions in HCl, Na-and Cl-ions in NaCl, and OH-and Na-ions in NaOH¹. As stated above, sodium chloride is chemotactically inactive, and from this it is evident that its components in solution—Na-and Cl-ions—give no action in this respect. It then follows directly that the attracting action of hydrochloric acid is due to the H-ions, and the repellent action of sodium hydroxide to the OH-ions. From this

1. These agents are practically completely dissociated at 1/1000 mol. It is certain that their strengths at the diffusion-zone a little removed from the mouth of a tube containing 1/200 mol, for instance, are less than 1/1000 mol, by which the spores may be first stimulated.

standpoint the weak action of boric and hydrocyanic acids must be ascribed to their less dissociable character, that is, to the small amount of the H-ions present in their solutions. It is scarcely possible in the present case to find any evidence for the attraction of undissociated molecules, though they are known in some cases to exert chemotactic action on some organisms, such as the spermatozoids of mosses (PFEFFER, '84, p. 430), Bacteria (PFEFFER, '88, p. 604-605), spermatozoids of *Marchantia* (LIDFORSS, '05), etc. When we bear in mind that the attraction and repulsion accord well with the intensity of acidity and alkalinity of the solutions, it can scarcely be doubted that the H- and OH-ions are the principal active agents in the cases under discussion.

Referring to STANGE's paper, we thus find at once that we have reached a conclusion directly opposed to him regarding the action of acids upon the swarm-spores of Myxomycetes—the same species being the object of the experiments. Although he did not express any view as to the attracting component in the solution of the acids which he found to act positively, we may say that it is a natural consequence of his results that the anions must be regarded as the active agents in his experiments, while according to our results the cations are so. After all, therefore, we find that the swarm-spores of Myxomycetes show in chemotaxis a parallel phenomenon with certain Infusoria (JENNINGS, '97, JENNINGS and MOORE, '02), inasmuch as they respond to the H- and OH-ions.

While thus the swarm-spores respond very plainly to the chemotactic stimulus of the H-ions, it may be mentioned that their reactions to several acidic substances are most remarkably divergent, a fact apparently inconsistent with our present conclusion. The first thing to be noted on this point is that the attraction by inorganic and organic acids is not necessarily in accordance with their "isohydric solutions," some being repellent and others attractive although isohydric with each other: in short, the intensity of attraction does not accord accurately with the amount of the H-ions present in the solutions. GARREY has also noticed the occurrence of such phenomenon in the reactions of *Chilomonas* to organic and inorganic acids. He says, "while inorganic acids show the

same effects if they contain the same number of H-ions in the unit volume of the solution, the organic acids do not show a definite relation between the number of H-ions and effects produced" (p. 309). In the next place, it must be remarked that while in weaker isomolecular solutions the acids and the acidic solutions of their salts have similar chemotactic actions on the spores, their effects are different if the solutions are stronger: the acids prevent the approach of the spores to the diffusion-centre, while their salts do not. These facts give us a hint on the existence of any other component which may give a different action from that of the H-ions in the given agents. As a rule, undissociated molecules must be present in solutions of less dissociable acids and salts, their number varying according to their dissociability, and their presence or absence, or their specific characters in different agents as regards the action on the spores would induce the above mentioned peculiarity. These points will be elucidated in later sections.

V. Motor Reaction in Chemotaxis.

In free-swimming organisms two different kinds of tactic movements may be distinguished, as already observed by previous authors. An extensive account of them is given by ROTHERT ('01), and PFEFFER ('04, p. 755) has proposed the names of topotaxis and phobotaxis for them. In topotactic movement the stimulated organisms orient their long axes parallel to the line of diffusion of the stimulant, and swim directly towards or away from the stimulant, according as the reaction is positive or negative. In phobotactic movement, however, the organisms react by reversing the direction of their motion, although the subsequent effect is the same as in the former case¹. The chemotactic reactions of the

1. In his recent elaborate work JENNINGS ('04) discusses at full length the motor reaction of unicellular animals to chemicals. Finding that their reactions are similar to what occurs in phobotaxis, he asserts that their behaviour is not in accordance with the tropism schema, the reaction being "motor reflex." PFEFFER, however, accepts, "Tropismus als Collectivbezeichnungen für alle physiologischen Reactionen" ('04, p. 755), and I will for the sake of convenience use the term phobotaxis in a wider sense.

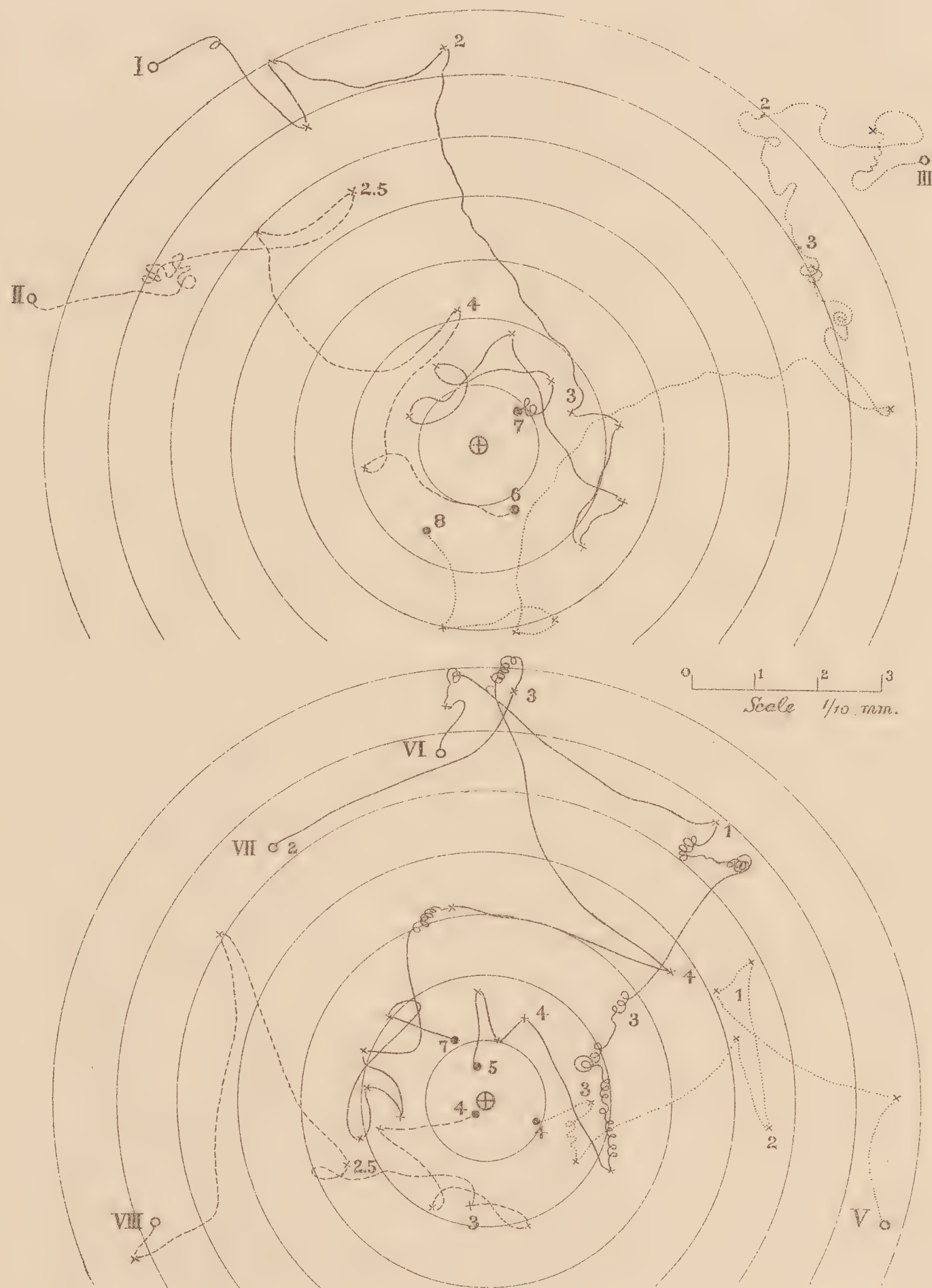
spermatozoids of many plants are known to be topotactic (PFEFFER, '04, SHIBATA, '05a), while Bacteria (ROTHERT, '01, JENNINGS and CROSBY, '02), Flagellata, and Infusoria (JENNINGS, '00, '02) react phobotactically. It is remarkable that the spermatozoids show a topotactic reaction only when it is positive; when the reaction is negative, they respond generally phobotactically (PFEFFER, '04, p. 759, SHIBATA, '05a).

As already known, with phobotactic organisms the difference between positive and negative taxis lies in the *motive cause of the stimulus* ("Reizanlass"), while the *reaction* is always the same, resulting in a change of the direction of motion (ROTHERT, '01, p. 400). We find, however, with topotactic organisms the reciprocal relation between the reaction and motive cause in giving positive and negative taxis. This means, as already remarked by ROTHERT, that with the former organisms increase in the intensity of the stimulus brings about negative, and its decrease positive taxis, while with the latter the tactic response is always produced by the increase in the intensity of the stimulus, negative taxis when the stimulus is strong and positive when it is weak. While on animals a most exhaustive study on phobotaxis has been made by JENNINGS ('97 and onward), in the plant kingdom there are no investigations which enable us to settle this problem. As a rule, the problem relating to tactic action must be very complicated and many points are obscure and hitherto untouched. At least, it may be said that while topotactic phenomena were exhaustively investigated recently by SHIBATA, no notable advance has been made in our knowledge on phobotactic phenomena since ROTHERT'S study, so far as the plants are concerned. For this reason I intend to describe as carefully as possible the conditions for attraction and in what manner the swarm-spores react in chemotaxis.

The collection of the swarm-spores in the capillary tube filled with an attracting substance takes place according to the typical phobotaxis scheme. As the spores move very slowly, I was able to observe accurately the mode of collection. The velocity, for instance, in *Aethalium*, which is the most active of the species studied by me, is on an average 0.01-0.016, mm. per second at a temperature of about 25°C. It is, therefore, very

easy to trace their path under the microscope with a pencil on paper, with the aid of a camera lucida.

In this way I could figure the motor paths of some swarm-spores of *Aethalium*, which were swimming about near the mouth of the tube and were being attracted to it, as shown in Text-fig. 1. The capillary tube with a calibre of 0.1 mm. contained 1/15-1/20 mol malic acid, and I fixed my attention in each instance on a very active spore swimming, at the time the tube is prepared, about 0.6 mm. from the mouth of the tube. After a minute it appeared to have been stimulated by the diffusing acid, and penetrated more and more into the inner diffusion-zone, until after a few minutes it reached just in front of the mouth from which the acid was diffusing out. At this moment a dense cloud had been formed at the mouth of the tube. As may be seen from the figure, the movement was continuous throughout the inner diffusion-zones and the spore appears to be quite indifferent to the action of the acid. But if it happened to pass out from a certain zone in which it had been swimming into an outer zone, i.e., the zone with less amount of the acid, reaction was observed instantaneously: it stopped suddenly, came to rest for a while, then turning the anterior end of the body on one side oriented the long axis of the body in some other direction than the one it had been taking, and renewed its active forward motion just in a startling manner. In this way the spore could not get out of the zone in which it was swimming. Whenever it came out into an outer zone this reversal of motion occurred invariably. The point where a transitory interruption of the motion or an alteration of its direction takes place may be determined by noting its distance from the innermost zone into which the spore had originally penetrated: in the present case, it was found to be 0.05-0.1 mm. in round number. This means that the spore is sensitive to a decrease in the amount of the acid denoted by the distance of the two zones. In this connection it must be remarked that the spore can easily accommodate itself to the concentration of the acid in which it lies, and when it has penetrated into a deeper zone, the point for the reversal of motion lies nearer the source of the stimulus, thus leading the spore to reach the optimal zone.



Text-fig. 1. Motor paths of swarm-spores of *Aethalium* reacting to malic acid $1/20$ mol in a capillary tube 0.1 mm. in diameter. Roman numerals give the number of observations; Arabic numerals in each course give the time in minutes after the

insertion of the tube; \odot , position of the spore at the beginning of the observation; \times , interruption of the movement; \bullet , position of the spore at the end of the observation; \oplus , centre of the diffusion-zone. (Drawn with the aid of a camera lucida. Zeiss 2 \times B.)

From the above it will be seen at once that all equally stimulated spores do not necessarily reach the centre of the stimulus with equal ease. For, those spores whose motion was directed towards the source of stimulus, i.e., towards the mouth of the tube in the present case, would directly reach the inner diffusion-zone, and the first reversal of motion would take place somewhere near the centre of the stimulus. In this case only a few reversals of motion would be sufficient to bring the spore to the desired zone of diffusion. If, on the other hand, a spore should be swimming in a direction not parallel with the line of diffusion, it would require repeated reversals of motion to bring the direction of its motion towards the centre of the stimulus. Consequently, some spores would have to traverse a long path with repeated reversals of motion, before coming to the zone of equilibrium, while others at equal distances from the source of the stimulus might reach the same zone more quickly. According as the path is long or short, the time required would vary greatly. This is clear at a glance on the figure given above.

As in phobotaxis the spores react to a decrease of the stimulating substance, it is quite obvious that their collection in a certain zone depends upon several factors. Firstly, for a dense collection to be formed the stimulating zone must be large enough to give stimulus to numerous spores distributed in wide area. This may be seen by comparing the effects of larger and smaller tubes, both containing equimolecular solutions of a given agent. Secondly, since the reversal of motion is due to a certain decrease in the amount of the stimulating agent at the turning point, it is necessary for a good collection of spores that two adjacent zones should present a large difference in the amount of the stimulating agent. For, in this case the spores moving from an inner to an outer zone will come to feel the decrease of the stimulant sooner than when the difference is smaller. Consequently, reversals of motion would take place more

frequently in a unit time and the spores would reach the zone with a more concentrated solution of the agent sooner. In my opinion, the diffusion-zones are under this condition when the tube contains a comparatively strong solution of the agent. It may be due to this that a tube with 1/15-1/20 mol malic acid attracts the spores more densely than one containing 1/50-1/100 mol. Of course, it must be noted that in the latter the outermost zone which causes the first reversal of motion lies nearer to the mouth of the tube than in the former, and consequently the number of the spores that are acted on by the acid must be less. However, the difference in the density of the collection near the mouth of the tube mentioned above is not due to this cause, but depends in all probability upon the condition of the diffusion-zones quoted above.

As a general rule, a dense collection of organisms at the mouth of the capillary tube once formed becomes gradually diffuse and faint later. This was very clear in my experiments. The chief cause of this change is very likely a gradual decrease in the difference of the amount of the stimulating agent at successive diffusion-zones. In fact, the decrease may proceed so far that an approximately uniform distribution of the agent is arrived at in all the diffusion-zones.

ROTHERT has remarked that a collection is formed more rapidly the more swiftly the organisms swim. This is particularly true in the case of phototaxis. In this connection it may be mentioned that the swarm-spores of *Aethalium* require, when the tube contains 1/15-1/20 mol malic acid, 5-10 minutes to attain to their final position from the point where the first reversal of motion occurs. Other things being equal, the much slower spores of *Stemonitis* and *Comatricha* require much more time for the same process.

It is to be noted that in slowly moving, phototactic organisms an unfavourable condition of the diffusion-zones may come about before the reactions are gone through. As stated above, the difference in the amount of the stimulant in two successive zones becomes less and less with the lapse of time, or before the arrival of the organisms in these zones, and the backward motion becomes less easy than in an earlier condition. In ac-

cordance with this view I could observe that the collection of the slow spores of *Stemonitis* and *Comatricha* is not so conspicuous as in *Aethalium*, although they were brought under the action of the same stimulant under the same condition, the condition of the diffusion-zones becoming certainly unfavourable when the spores approached the stimulating zone.

In phobotactic reactions it must be noted that the entry of the organisms into the tube is by no means a necessary effect. For, differently from topotaxis, the stimulated organisms do not orientate themselves necessarily towards the mouth of the tube from which the stimulant is being diffused out, or, in other words, they have no ability, so to speak, to find out, as in the topotactic reactions, the direction of diffusion or the source of the stimulant. The entry into the tube is effected only when the direction of the final motion is parallel to the line of diffusion. This, however, is due to chance, so that some spores may take this direction after a few reversals of motion, but for most spores, there will be many reversals before the entry is effected. Such being the case, it is evident that a tube with a larger diameter gives much more chances for the entry of the spores than a smaller one.

A small tube not only does not show the definite effect of phobotaxis, i.e. capture of the spores in the tube to best advantage, but its action is also weak owing to the small optimal diffusion-zone and of the small difference in the amount of stimulant at successive diffusion-zones. I am now therefore strongly inclined to think that the divergent results of STANGE's and mine may be ascribed to the different size of the capillary tubes we used. He used a tube less than 0.02 mm. in diameter (13-15 micromillimetre). A tube of this size does not bring forth positive effects on such a slowly swimming organism as the swarm-spore of *Myxomycetes*. His failure in finding out the positive action of many acids may reasonably be ascribed to this defect in his methods of experiment. The following table shows the different effects of the diameters of the tube on the chemotactic movement of the swarm-spores of *Aethalium*:

Table III.

Diameter of the tube filled with 1/20 mol monopotassium phosphate.	Time elapsed after pre- paration.	Result.
0.24 mm.	10 minutes	Remarkable collection.
0.15 "	10 "	"
0.15 "	5 "	Collection diffuse round the mouth.
0.1 "	10 "	Faint collection with very few spores.
0.09 "	5 "	Collection not apparent; few spores round the mouth.
0.05 "	10 "	No collection.
0.03 "	5 "	"
0.02 "	10 "	"
0.01 "	5 "	"

After all, it can be said with safety that in chemotactic experiments with capillary tubes on the swarm-spores of Myxomycetes, the conditions of diffusion-zones round the mouth of the tube must be taken first of all into consideration in order to obtain accurate results. The conditions vary, as already stated, with the time of observations, the concentration of the substances in the tube, and the diameter of the tube. These conditions have a closer connection with phobotactic than with topotactic reaction.

The reactions in negative phobotaxis is quite similar. That positively phobotactic organisms may be also negatively phobotactic was established by ROTHERT ('01, p. 394) and conjointly by JENNINGS and CROSBY ('02, p. 35). As quoted above, an organism showing positive topotaxis may also show negative phobotaxis (PFEFFER, '04, p. 759, SHIBATA, '05a, p. 609). Hence, negative phobotaxis is a universal phenomenon among chemotactic organisms. Now, when the swarm-spores of Myxomycetes are acted on by a repellent substance, we observe

that as soon as they approach a zone in which the substances are contained in sufficient amount to stimulate, they stop suddenly, come to rest for a while, and then move away quickly in an arbitrary direction. The manifestation of this repulsion is most apparent when the spores are exposed to the action of strong acids. The repellent component in this case is doubtless the H-ion. The spores are first attracted by the given acids and arrive at the optimal diffusion-zone. They are thus brought close to the supraoptimal zone which lies inside the optimal zone, and also to the infraoptimal zone lying just outside. When the spores freely swimming in the optimal zone may happen to pass out into the supra- and infraoptimal zones, the reaction is expressed invariably by the reversal of motion.

Some remarks may be made on the action of acids upon the movement of some other organisms. As regards the reaction of *Chilomonas* to several acids GARREY ('00) distinguished two causes for their collection in a certain zone of diffusion, viz. *chemokinesis* ("Unterschiedsempfindlichkeit") and positive chemotropism. This view is, however, criticised by JENNINGS and MOORE ('02), who observed that the given animal responded to acids in the form of phobo-chemotaxis (according to PFEFFER) or of a "motor reflex" (according to JENNINGS' terminology). In *Myxomycetes* the facts thus far obtained lead me to agree with JENNINGS' ('00a) view.

Lastly we will consider the nature of phobotactic reactions. It is true that, strictly speaking, phobotactic reactions in the cases considered above are not tropistic, as maintained by JENNINGS ('04). We have to do with tropism when a reaction shows a direct relation to the direction of application of the stimulus (PFEFFER, '04, p. 83, 547), that is, to the directive movement. In phobotaxis, however, the orientation of the organ or organism does not show any definite relation to it, the reaction taking place by backing whenever a stimulus is felt. JENNINGS found in lower unicellular animals that they turn, when stimulated, towards a structurally defined side, wherever the stimulus may impinge. This leads us to recognize a close resemblance between phobotactic and nastic movements

(PFEFFER, '04, p. 83, 356). JOST ('04, p. 673) has already expressed the same view. However, the phobotactic response of Bacteria to chemical stimuli is manifested by a backward movement, with the posterior end foremost (ROTHERT, '01, JENNINGS and CROSBY, '02). Since in this case it is difficult to distinguish the structurally defined side, and the motion is different from that of the lower animals quoted above, we can not compare it with nastic movement. Now, since it is difficult to distinguish a structurally defined side in the swarm-spores of Myxomycetes just as in Bacteria, we can not decide whether the reaction is similar to what takes place in the nastic movement or not. The essential difference in the motive cause of the phobotaxis and topotaxis is indeed a most difficult question. According to ROTHERT and PFEFFER topotactic chemotaxis is due to the differential distribution of the stimulant on different sides of the organism, while in phobotactic chemotaxis the differential concentration of the stimulant at the two poles of the organisms is concerned. When we recall, however, that topotactic spermatozooids mostly rotate on their own axes, it appears very improbable, as JOST remarked (his Lecture, English edition, 1907, p. 545), that the inequality of the stimulus on opposite sides is the motive cause of topotaxis as distinguished from phobotaxis. At present, we can only say with JENNINGS ('04, p. 111) that the alteration of the direction of motion depends upon a very complicated physiological state involved in the body of the organism.

VI. Ring-Collection.

When a capillary tube of 0.1 mm. inner diameter filled with 1 mol sulphuric acid is inserted in the culture-medium of the swarm-spores of *Aethalium* under a cover-glass, we will observe that the spores react immediately and gradually approach the source of the stimulant. After 10 minutes an immense number of the spores have entered a diffusion-zone whose diameter is 4-6 mm. These spores, however, do not penetrate

further owing to the repellent action of the acid. So that they now form a very distinct ring, 0.2-0.25 mm. in thickness, which can be easily recognized with the naked eye as a white stripe by means of reflected light, and remains unchanged for an hour.

The ring-collection seems to be a general occurrence in tactic organisms whatever the kind of stimuli may be. Instances of it have been enumerated by ROTHERT ('01, p. 402), on the basis of which we may distinguish two different causes according as the stimulus is furnished by dissociable or undissociable agents. As a rule, in the tactic reaction to light, heat, oxygen and other undissociable chemical agents, the organisms assemble from all sides to a zone where the optimal amount of the stimulant is contained. But when dissociable agents are used as stimulant, for instance, in chemotaxis, different components such as anions, cations, and free molecules, may come into play, and may exert different stimuli. If in this case two or more components have antagonistic actions, the organisms will assemble in a zone where both actions are just neutralized (PFEFFER, '04, p. 811). According to BULLER ('00, p. 567) the ring-collection of fern-spermatozoids with strong solutions of malic acid is effected by the attraction of the anion and the repulsion of the cation (H-ion). A similar view was announced by SHIBATA in his experiments on the spermatozoids of *Isoetes* ('05a), *Salvinia* ('05b), and *Equisetum* ('05c).

To ascertain the cause of the ring-collection in the present case we must first find out the components contained in the given solution of sulphuric acid and their chemotactic action upon the spores. There is no doubt that free molecules, anions, and cations are present in different amounts, and since we have seen in the foregoing experiments that the cations attract the spores when in right strength, we must now attempt to find whether the cations, anions, or free molecules repel the spores in the strength obtaining in the given zone. Before going into the discussion of the problem, it will be well to consider minutely the relation between the ring-collection and the characters of several weak and strong acids, and in particular to study the character of the ring formed in each

acid.

(a) Thickness of the ring and the character of its margins.

The ring formation in other acids is essentially similar to that in sulphuric acid. Its diameter and thickness however vary according to acids, even in equimolecular strengths. Precisely similar rings are obtained with hydrochloric, nitric, and phosphoric acids. In these the inner and outer boundaries of the ring are sharply defined, the number of spores decreasing suddenly outside them. Of organic acids, oxalic induces a nearly similar ring as the inorganic acids just mentioned, that is, the ring is very large and has distinct margins, though the thickness is somewhat greater, being 0.3-0.4 mm. In acetic acid the ring is slightly smaller than in sulphuric, hydrochloric, and nitric acids, and the number of the spores in the ring is less, both the outer and inner margins are indistinct, and its thickness measures 0.3-0.4 mm. In tartaric acid is formed a most obscure, small, and very thick ring with indistinct margins. Other acids induce more or less characteristic rings. Arranging all the acids tested in the order of the characters noted above, we get the following result (Table IV):

Table IV.

Thin and clear ring in descending order.	Thick and diffuse ring in descending order.
Sulphuric acid.	Tartaric acid.
Phosphoric acid.	Chromic acid.
Hydrochloric acid.	Malic acid.
Nitric acid.	Citric acid.
Oxalic acid.	Lactic acid.
Formic acid.	Acetic acid.

Thus the thickness of the ring increases and the distinctness of the margins decreases from sulphuric towards formic acid, and further from formic towards acetic acid until they attain the maximum and minimum respectively in tartaric acid.

Generally speaking, in inorganic acids the ring is mathematically circular and the spores are distributed uniformly throughout the ring. But in certain organic acids, such as citric, tartaric and lactic, the ring is frequently seen to be irregular, and the spores aggregate more densely at certain points.

(b) Size of the ring.

The diameter of the ring varies even with the same acid according to the diameter of the capillary tube. When a comparatively large tube (0.13-0.14 mm.) is used, the amount of the diffusing acid is naturally large, and the optimal zone for the collection of the spores may have a large diameter. The size of the optimal zone also varies according to the duration of diffusion, increasing in some acids and decreasing in others. At one time I have observed with a tube (0.13 mm. diam.) filled with 1 mol sulphuric acid a ring of 8 mm. in diameter 30 minutes after the insertion of the tube. But in one and a half hours from the beginning, the diameter has increased to 11 mm. With other acids, such as phosphoric, malic, citric, tartaric, etc., the ring decreases in diameter, the effect being most marked after 1-1.5 hours.

Under the same condition different acids give rise to rings of different diameters. Repeated experiments have shown that mineral acids in general and some organic acids, for instance, oxalic acid, produce larger rings, while the other organic acids, as malic, lactic, acetic, and tartaric, attract the spores in smaller rings. For the sake of convenience, I may divide the acids used into four orders, according to the size of the ring formed, viz. (1) sulphuric, hydrochloric, nitric, and chromic acids, inducing the largest rings, (2) oxalic and acetic acids the next large rings, (3) citric, malic, lactic, and formic acids small rings, and (4) tartaric acid the smallest ring. The following table shows approximately the relative size of the rings with various acids (1 mol) in tubes of different diameters and after different time intervals of observation.

Table V.¹

Acid.	Diameter of the tube (mm.).	Size of the ring at first observation.		Size of the ring at second observation.	
		Inner diameter (mm.). ²	Time (min.).	Inner diameter (mm.).	Time (min.).
Sulphuric acid	0.11	5.0	20		
" "	"	5.0	10	7.0	25
" "	"	4.0	10	5.0	40
" "	0.1	4.0	20	6.0	40
Hydrochloric acid	0.12	4.0	10	5.0	40
" "	0.11	5.0	10	6.0	25
Nitric acid	0.13	4.4	10	6.0	40
" "	0.07	3.4	10	4.0	25
Chromic acid	0.11	5.0	20		
" "	0.12	6.0	20		
Acetic acid	0.15	4.0	10	4.0	20
" "	0.14	4.4	20	3.0	90 ³
Malic acid	0.12	{2.0 4.0*	20		
" "	"	3.0	10	{3.0 4.0*	20
" "	0.07	3.4	10	{2.0 5.0*	30
Oxalic acid	0.13	4.0	20	5.4	60
" "	0.1	2.5	20	{2.0 2.6*	40
Citric acid	0.15	4.0	10	{4.0 5.0*	20
" "	0.14	3.5	20	{4.8 5.4*	60
" "	"	4.0	20		
Lactic acid	0.15	3.0	15		
" "	0.12	3.0	20		
Formic acid	0.12	3.5	10		
" "	0.11	3.0	10		
Tartaric acid... ..	0.15	{3.0 3.5*	10	{2.0 5.0*	30
" "	?	{1.0 3.5*	20		
" "	?	{1.0 2.4*	20		

1. The outer diameter of the ring is marked with an asterisk.

2. The diameter of the repulsion-space.

3. Motionless spores congregate at the inner margin of the ring and make the repulsion-space gradually smaller.

This table shows that the rings formed in mineral acids are all similar to that in sulphuric acid: the sizes are nearly equal and the margins very distinct. As we know, these acids are highly dissociable. It is, therefore, certain that the ring-zone contains the optimal amount of the H-ions; at the inner zones they are supraoptimal and at the outer infraoptimal. From several other experiments we may say that the other components in the solution, namely acid radicals and free molecules, have no action upon the spores in the amount they are present at the outermost repulsion-zone. Or, if they exert any stimulus, it has no influence upon the formation of the ring.

Now with organic acids. Unless the dissociable condition of these acids is not considered, we can hardly form any appropriate explanation of the ring-collection. Since they are mostly weak, the amount of the H-ions produced in equimolecular concentrations with inorganic acids must be necessarily less. When we consider that the repulsion-space is nevertheless disproportionately large compared with the dissociability of each acid, we should have to conclude that there must be a component or components acting repellantly on the spores in a certain concentration. Remarkable is the repulsion-space in acetic acid. It is perhaps the largest to be formed in organic acids, except oxalic in which, owing to its strong acidity, the repulsion-space may be as rich in the H-ions as in mineral acids. It is easy to imagine that the H-ions are here far from being optimal in amount in the ring-zone and that some repulsive components coexist prevent the approach of the spores towards the optimal zone lying nearer to the mouth of the tube. The following table will show at once how small is the degree of dissociation of acetic acid as compared with hydrochloric acid (see OSTWALD, '89, p. 174):

Table VI.

Concentration in mol.	Amount of H-ions per litre.		Relative amount of H-ions in both acids.
	Hydrochloric acid ¹	Acetic acid.	
1/8	0.125	0.00148	84.5 : 1
1/16	0.0625	0.00104	60.0 : 1
1/32	0.0312	0.00074	40.0 : 1
1/64	0.0165	0.00052	30.0 : 1
1/128	0.0078	0.00036	21.6 : 1
1/256	0.0039	0.000256	15.2 : 1
1/512	0.00195	0.000173	10.9 : 1

Thus we see that in the amount of the H-ions 1/8 mol acetic acid corresponds nearly to 1/512 mol hydrochloric acid. Hence, were the repulsion-space in acetic acid is simply due, as in hydrochloric acid, to the supraoptimal amount of the H-ions, it should be considerably smaller. The case, however, being otherwise, it is certain that there is a large predominance of undissociated molecules at a certain zone, exerting a repellent action upon the spores attracted to it by the H-ions. Therefore, as a matter of fact the zone where the spores form a ring would not contain the optimal amount of the H-ions, but the attraction of the H-ions and the repulsion of the free molecules are in a state of equilibrium in it. That the collection of the spores in the ring is not so dense as in the case of hydrochloric acid is an additional proof for this view.

In the case of the ring-collection in other acids the same consideration must be taken into account. The size of the repulsion-space accords in general with the degree of dissociation, though the specific character of the molecules of each acid may come into play—a subject on which I am not ready to say anything definite. Hence, in studying the character of the ring formed in these acids a comparison of their dissociability is indispensable. This is shown in the following table (OSTWALD, '89, p. 418-422, WALDEN, '92, p. 568, NERNST, '00, p. 469):

1. Here it is assumed to dissociate completely at about 1/8 and 1/16 mol.

Table VII.

Acid.	Affinity coefficient, 100 k.
Butyric acid	0.00149
Acetic acid	0.00180
Succinic acid	0.00665
Lactic acid	0.0138
Formic acid... ..	0.0214
Malic acid	0.0395
Citric acid	0.082
Tartaric acid	0.097
Salicylic acid	0.102
Oxalic acid	10.0 (?)

If we now compare this table with Tables IV and V, the relation between the acids and the size as well as the thickness of the ring will be easily seen. For instance, tartaric acid is more dissociable than many other organic acids. This induces the spores to approach the inner diffusion-zones much nearer owing to the attraction of the H-ions and offers less hindrance to their approach owing to the lesser amount of the free molecules than in the less dissociable acids, like malic, formic, and lactic. We must also ascribe to the same cause the greater thickness of the ring in tartaric than in the acids just mentioned. Again, the larger size of the repulsion-space in acetic than in any other organic acid is doubtless due to the presence of more numerous undissociated molecules.

It is very interesting to compare in this place my results with those obtained by GARREY in his experiments with *Chilomonas* as regards the formation of the repulsion-space in several inorganic as well as organic acids. He gave the least value of the solution of acids as forming the repulsion-space (clear space according to him) as follows:

Hydrochloric acid	1/1000-1/1200 mol.
Nitric acid	"
Sulphuric acid	1/2000-1/2400 mol (N/1000-N/1200).
Oxalic acid... ..	1/60 mol (N/300).
Tartaric acid	1/600 mol (N/300).

Formic acid	1/100 mol.
Lactic acid...	1/1000 mol.
Acetic acid...	1/200-1/300 mol.

GARREY calculated the amount of the H-ions in these solutions (p. 308): oxalic acid contains three times as much as 1/1000-1/1200 mol hydrochloric acid, tartaric acid equal amount, formic acid three times, lactic acid less than one third, and acetic acid one third as many H-ions, each in the given strength.

It is, therefore, plain that the given solutions are not isohydric. About this singular fact he did not give a satisfactory explanation, saying only, "In some case, *e.g.*, that of acetic acid, the effects are greater than could be expected from the degree of dissociation" (p. 315). And referring to RICHARDS and KAHLENBERG's view he adds, "By comparison with inorganic acids the sour taste of acetic acid was about three times as intense as the number of H ions warranted" (p. 309). Thus he seems to think that the relative action of the organic acids quoted above is due to the specific behaviour of each acid independently of the number of the H-ions present, but he did not pay attention to the action of other components that might be present. To ascertain whether the anions or the free molecules are concerned in the formation of that repulsion-space further investigation is desirable. In my case the fact that the molecules repel the spores is hardly to be denied, the intensity of repulsion being different in different acids. If the same thing applies to GARREY's experiments, we can say that the relative intensity of the repellent action of the molecules of organic acids upon *Chilomonas* is not parallel to that upon *Myxomycetes*. For instance, lactic acid acts more strongly than formic acid on *Chilomonas*, while both acids act equally on *Myxomycetes*.

In the next place, there exists certain relation between the thickness of the ring and the distribution of the H-ions in the diffusion-zones, latter being different in different acids. Generally speaking, in strong inorganic acids a thin ring with distinct margin is formed, while in many organic acids the ring is thick and the margin indistinct, the spores in the ring being also less numerous. It is evident that in the former

the optimal number of the H-ions is present within a narrower limit. Taking for example, hydrochloric and acetic acids for comparison, one may easily understand that the decrease in the number of the H-ions at the successive diffusion-zones is very gradual in acetic acid. If we admit that both acids diffuse with equal velocity, we shall have equal concentration of them at a given zone. Assume that this zone contains $1/64$ mol of the given acids. Then the difference of the number of the H-ions between that zone and that just preceding or succeeding it will be, as may be seen from Table VI, greater in hydrochloric than in acetic acid: the H-ions amount in the former acid to approximately 0.0156 and in the latter to 0.0052 gram. per litre, while in the next outer zone ($1/128$ mol) they decrease to 0.0078 and 0.0036 respectively, that is, in hydrochloric acid they decrease to one half and in acetic acid to two thirds. A similar relation may be obtained at any other zone. From this we may conclude at once that the extent of the zone which contains the optimal number of the H-ions for the collection of the spores should in hydrochloric acid be two thirds as great as in acetic acid; for instance, if 0.00025-0.000125 gram. per litre of the H-ions ($1/4000$ - $1/8000$ mol solution of acid) be assumed as the optimal amount in hydrochloric acid, the width of the zone which contains the same amount of the H-ions in acetic acid will be 1.5 times. The same argument may be applied to all other weak acids. In consequence, the spores remain more scattered in weak than in strong acids, so that the margin of the ring is more or less indistinct in the latter.

In the last place, the relative thickness of the ring and the distinctness of its margin will depend upon the valency of acid radicals, and also upon the relative amount of the H-ions and free molecules. It is clear that at equal distances from the mouth of the tube dibasic acids evolve much more H-ions than monobasic acids, and tribasic acids more than dibasic acids, provided they be all equally dissociable. So that the H-ions will be distributed in sufficient amount to exercise attraction in a wider area in tribasic than in dibasic acids, and in dibasic than in monobasic acids. This view is supported by the fact that citric, tartaric,

and malic acids give thicker rings than formic, lactic, and succinic acids.

When the free molecules of acids come into play in forming the ring, we must consider the relative amount of the H-ions and molecules at successive diffusion-zones, which may differ in different acids. The thickness of the ring is determined by the difference of radii of the attraction-space by the H-ions and the repulsion-space by the molecules, the larger the difference the thicker the ring. The somewhat thinner ring formed in acetic acid may be ascribed to the small difference of the two radii. On the other hand, the thicker ring formed in the more dissociable malic and tartaric acids is certainly due to the larger difference of them, the radius of the repulsion-space being much shorter than that of the attraction-space, a fact easily comprehensible when we consider that they are dibasic and more easily dissociable than acetic acid.

From the foregoing observations we may make the following statement regarding the ring-collection:

The cause of the ring-collection is different in *strong* and *weak* acids, or in other words, according as they are more or less easily dissociable. In strong acids it is associated with the amount of the H-ions only, while in weak acids it is a resultant of the attraction by the H-ions and the repulsion by the free molecules. In the latter case the diffusion-zone where the spores form the ring does not generally contain the optimal amount of the H-ions; on the other hand it is more or less infraoptimal, the coexisting molecules preventing their penetration into the inner zone having the optimal number of the H-ions. The character of the ring formed in the weak acids are associated with the dissociability and valency of acids, and with the relative length of the radius of the attraction-space of the H-ions and the repulsion-space of the free molecules, etc. This conclusion will be sufficiently confirmed by the experiments to be described in the next section.

VII. Column-Collection.

When a capillary tube filled with more dilute solution of acids than was used in the preceding experiment, for instance, 1/200-1/100 mol of certain organic as well as inorganic acids (HCl , HNO_3 , H_2SO_4 , malic, acetic, formic, lactic acids, etc.) is used in the experiment, the swarm-spores come immediately to approach the mouth of the tube. After 10-15 minutes they form a dense collection at the mouth, and still later, advance into the tube forming a cloudy column of various length, but mostly of 0.3-0.5 mm. At first the column lies near the mouth, but sooner or later according to the strength of the solution the column advances as a whole deeper into the tube. In one case there was a displacement of as much as 2 mm. in one hour. In 1/200 mol sulphuric acid the displacement was hardly noticeable, and in 1/100 mol hydrochloric, nitric, malic, formic, and acetic acids there was a displacement of 0.5 mm. in 40-60 minutes, but when 1/200 mol of these acids and 1/400 mol sulphuric acid were used, the displacement was 1-2 mm. or more in one hour.

The length of the column may vary according to different acids and different concentrations of the same acid. 1/200 mol hydrochloric and oxalic acids and 1/400 mol sulphuric acid give rise to a column of 0.4 mm. in length, while in 1/200 mol acetic and malic acids it is about 0.5 mm. After 50 minutes the column given by 1/200 mol malic acid attains the length of 1.5 mm., while by its 1/100 mol it is only 0.5 mm. long after the same interval of time.

The column-collection, as in the ring-collection, must be due to the presence of the optimal concentration of the H-ions or to the equilibrium of the attraction by the H-ions and the repulsion by the molecules of acids. In either case the zone at which the spores are assembling is wider here in extent than in the ring-collection. This is evidently connected with the velocity of the diffusion of the agent; in the tube the concentration of the agent at each successive diffusion-zone decreases much less quickly than outside the tube, and in this case the optimal amount

of the stimulant is contained in a wider space than in the case of the ring-collection.

When the columnar collection of the spores in the tube becomes apparent, scarcely any spore is to be found outside. Certainly, the solution near the mouth has fallen by this time below the critical concentration for exerting a sufficient stimulus, while the zone of optimal concentration is gradually displaced deeper into the tube. The rapidity of displacement is not all equal in different concentrations of acids contained in the tube and also to the different kinds of acids to be tested, as well be seen from the following table:

Table VIII.

Acid.	Concentration (mol).	Diameter of the tube (mm.).	First observation.			Second observation.		
			Distance of the outer extremity of the column from the mouth.	Length of the column (mm.).	Time observed (min.)	Distance of the outer extremity of the column from the mouth.	Length of the column (mm.).	Time observed (min.).
Sulphuric acid ...	1/200	0.16	0.0		15	0.0		60
" " ...	"	0.2	0.3		75			
" " ...	1/400	0.15	1.5		20	1.5		60
" " ...	"	0.25	0.5		15	0.7		60
" " ...	"	0.17	0.7		30	1.0		60
" " ...	1/600	0.2	1.5	0.6	40			
" " ...	"	0.17	2.0		30			
Hydrochloric acid	1/100	0.11	0.0		10	0.1		25
" " ...	1/200	0.2	1.0		75			
" " ...	"	"	0.3	0.4	10	0.4		40
Nitric acid ...	1/100	0.1	0.0		10	0.2		25
" " ...	"	?	0.0	0.0	10	0.2	0.3	50
" " ...	1/150	0.13	0.2		20	0.3		90
" " ...	"	"	0.1		10	0.5		60
" " ...	1/200	0.13	0.1		10	2.0		60
" " ...	"	0.15	0.5		20	0.5		60
Caromic acid ...	1/200	0.1	0.0	0.5	10	0.5	0.5	30
" " ...	1/300	0.13	0.0	1.5	30			
" " ...	"	?	0.5	0.4	90			

Acid.	Concentration (mol.).	Diameter of the tube (mm.).	First observation.			Second observation.		
			Distance of the outer extremity of the col- umn from the mouth.	Length of the column (mm.).	Time observed (min.).	Distance of the outer extremity of the col- umn from the mouth.	Length of the column (mm.).	Time observed (min.).
Chromic acid ...	1/3000	0.15	1.0	1.0	10	2.0	1.0	30
Picric acid ...	1/100	0.1	0.0	0.7	20	0.5		50
" " ...	1/200	0.14	0.0	0.7	20	0.2	0.5	50
" " ...	"	0.11	0.5	0.5	20	1.0	1.0	50
" " ...	"	0.15	0.2	0.4	15	0.5	0.5	50
" " ...	1/300	0.13	0.6	0.5	20	1.0	0.5	50
Oxalic acid...	1/100	0.15	0.0		10	0.0		50
" " ...	1/200	0.2	1.0		75			
" " ...	"	0.15	0.0		15			
" " ...	"	"	0.0		10	0.3		60
" " ...	"	0.13	0.0		20	0.0		96
Formic acid ...	1/100	0.17	0.2	0.3	40			
" " ...	"	0.14	0.3	0.5	35			
" " ...	"	0.12	0.5		10	0.8	0.4	50
" " ...	1/200	0.2	1.3	0.5	35			
" " ...	"	0.17	1.0		15	1.0		60
" " ...	"	0.2	0.5		15	1.0		60
Malic acid ...	1/100	0.15	0.0		20	0.3		50
" " ...	1/200	0.14	0.2	2.5	30			
" " ...	"	0.18	0.0		20	0.0	1.5	50
" " ...	1/300	0.17	0.0	3.5 (diffuse)	30			
" " ...	1/400	0.15	0.2	4.0 (diffuse)	50			
Lactic acid...	1/100	0.15	0.0		15	0.3		25
" " ...	"	?	0.2	0.7	40			
" " ...	1/200	0.15	?	?	30	1.0		45
Succinic acid ...	1/200	0.14	0.0	1.0 (diffuse)	10	0.3	3.0 (diffuse)	50
" " ...	1/300	0.13	0.2	1.0	10	1.0		40
Citric acid ...	1/200	0.16	0.5	0.5	15	0.5	1.0	50
" " ...	1/400	0.14	0.0	3.5 (diffuse)	15	1.0	5.0	50
" " ...	"	0.17	0.5	1.5	15	1.0	2.0	50
Tartaric acid ...	1/200	0.12	0.2	0.6	10	0.6	1.0	30
" " ...	1/400	0.14	0.5	1.0	10	0.5	5.0-6.0	35

As already described in the foregoing pages, the collection of the spores in the tube containing certain acidic salts presents somewhat different features. Both in weak and strong solutions the spores are distributed more diffusely and in a larger space. In one case I could observe the spores distributed uniformly from near the mouth to the extent of 5 mm. in the tube. It appears that there is no component counteracting the advance of the spores and that the stimulant is distributed more diffusely in the tube.

VIII. Chemotaxis in Mixtures of two Electrolytes Producing Common Ions.

According to the dissociation theory the degree of dissociation of a given substance undergoes a great change when it is mixed with another substance producing common ions (NERNST, '00, p. 471). Taking acetic acid for instance, its dissociability is diminished by addition of the more dissociable sodium acetate, as we see from the quantitative determination of ARRHENIUS ('90).

This fact seems to give us a hint for an explanation of the attraction of the H-ions and repulsion of the molecules. *A priori*, we may think that, if the H-ions are really attractive and undissociated molecules repellent, acetic acid mixed with its sodium salt would exert less attraction and stronger repulsion than if it acted alone under the same condition. This presumption is borne out by the following experiments:

Experiment 1.

Aethalium.

Tube *a* (diam. 0.13-0.12 mm.): acetic acid 1 mol.

Tube *b* (diam. 0.12 mm.): acetic acid 1 mol and sodium acetate 1/2 mol.

Tube *c* (diam. 0.12 mm.): sodium acetate 1/2 mol.

After 10 minutes we find in *a* a clear ring approximately of 3 mm.

in diameter and 0.3-0.35 mm. in breadth with dense collection of the spores. At the same time a ring of nearly similar dimensions is formed in *b*. It is, however, faint and diffuse, and the distribution of the spores in the ring is not uniform, showing weak attraction. In *c* no collection is formed.

After 35 minutes the ring of *a* becomes clearer but smaller with sharp inner margin, where rounded and motionless spores are found. Outside the ring active spores are still swimming about, so that the outer margin is not sharply delimited. In *b* the repulsion-space remains unchanged, but the ring appears to increase in thickness, both inner and outer margins being less definite, while the collection of the spores in the ring is not so conspicuous as in *a*. At the inner margin of the ring no remarkable accumulation of dead spores is in this case observable. Tube *c* does not attract any spores.

Experiment 2.

Aethalium.

Tube *a* (diam. 0.1): acetic acid 1/2 mol.

Tube *b* (diam. 0.1): acetic acid 1/2 mol and sodium acetate 1/2 mol.

Tube *c* (diam. 0.12): sodium acetate 1/2 mol.

After 10 minutes we find round the mouth of *a* a dense cloud of spores of 0.7 mm. in diameter and with no repulsion-space in its centre. The spores do not enter the tube, though they approach its mouth. In *b* is observed a very diffuse and faint cloud of nearly the same size. A clear space of 0.1 mm. diameter is formed round the mouth, so that there is a ring-formation of 0.3 mm. in thickness. *c* does not reveal any attraction or repulsion.

After 20 minutes no essential change is observable in the collection, except that the cloud becomes much denser in both *a* and *b*.

Experiment 3.

Aethalium.

Tube *a* (diam. 0.1): acetic acid 1/4 mol.

Tube *b* (diam. 0.1): acetic acid 1/4 mol and sodium acetate 1/4 mol.

A small cloud of spores is seen in both tubes, but in the latter the cloud is somewhat large, faint, and obscure owing to the comparatively small number of the spores.

These experiments show most clearly that the attraction of a mixture of an acid and its salt is much weaker than that of the acid alone. In all strengths a mixture gives a faint and diffuse collection. It may be due to the lesser amount of the H-ions at the diffusion-zone, or to small differences at the successive zones. Again a clear repulsion-space formed in the mixture may be ascribed to the larger amount of the acid molecules remaining undissociated. In the repulsion-space caused by the acid alone, the spores at the inner margin of the ring perish sooner or later, while the spores that are in contact with the repulsion-space of the mixture may escape this fate. It is evident that the difference in the two cases is associated with the different relative amount of the H-ions and acid molecules at the given zone. In pure acid the attraction of the H-ions is strong enough to attract the spores into the zone where the amount of the molecules is fatal to them, while in the mixture the same attraction is not so strong.

IX. Electrolysis and the Swarm-Spores.

When platinum wires of a dry battery of more than 1 volt are inserted as electrodes in a drop of the culture-medium of the spores on a slide, there will be attraction at the anode and repulsion at the cathode, and in a few minutes the spores collect along the wire of the anode, while those along the wire of the cathode show shrunken bodies and are either motionless or swim away phobotactically. When the strength of the current is doubled by using two batteries, a repulsion-space appears close to the wire of the anode, and a ring-formation is brought about. In this case small bubbles are often liberated from both poles.

The attraction and repulsion can still be observed after the circuit is opened. We see, therefore, that such reaction of the spores is not of galvanotactic nature, as galvanotaxis takes place only while the circuit is closed. It is highly probable that it is of chemotactic nature, due either to ions evolved by the electrolysis of certain electrolytes occurring in the culture-medium or to substances produced by the combination of the ions thus produced. The probable contents of the culture-medium may be a very dilute solution either of mineral salts when tap-water is used or carbonic acid in distilled or tap-water. Moreover, the calcium carbonate of the capillitia (in *Aethalium*) may come under consideration, as they may occur together with the spores in the culture-medium. These substances give rise to new substance. For instance, calcium carbonate is decomposed by the action of electric current into Ca and CO_2 . Both ions do not exist as such; Ca combines at the cathode with H_2O and produces H and $\text{Ca}(\text{OH})_2$, while CO_2 at the anode produces by a similar process O and CO_2H_2 . Thus as final products we obtain at the cathode a basic substance and at the anode an acidic substance, besides oxygen and hydrogen gas at the anode and cathode respectively. I have myself tested this result with litmus paper placed over the poles. Hence, there can be little doubt that the reaction of the spores must be due to the stimulus of the acidic and basic substances produced by the electric current. The acidic substance evolved at the anode is dissociated and accumulates the H -ions, while the basic substance at the cathode produces the OH -ions. When the current is strong, these ions become larger in amount at the respective poles, and act upon the spores in the manner already mentioned.

When a very weak solution of NaCl , $\text{Ca}(\text{CO}_3)_2$, or Na_2SO_4 is added to the culture-medium, the reaction of the spores becomes more conspicuous. On the other hand, if such medium is diluted with distilled water, the reaction is weakened. Such difference of reaction is certainly connected with the production of acid and hydroxide at the respective poles.

In the arrangement above mentioned we can not of course deduce any notion about the action of the galvanic stream, since no precaution

was taken about polarization, and the electrolysed substances may disturb the reaction of the spores to the given stream (PFEFFER, '04, p. 820). In fact, in passing the electric current of 1-2 volts no immediate reaction whether positive or negative is observable between the two poles, so long as the electrolysed substances are not yet produced. Should the spores be negatively galvanotactic, they should turn towards the cathode, which, as it evolves the OH-ions in the present case, must repel the approaching spores. Galvanotaxis has been ascertained in lower organisms, especially in Flagellata and Ciliata (VERWORN, '03). A special study of galvanotaxis of Myxomycetous swarm-spores will be carried on in future. At present, we must content ourselves with strengthening by electrolysis experiment the general conclusion that acidic substances attract and basic substances repel the spores.

X. Action of the Concentrated Solutions of Chemicals upon the Movement of the Swarm-Spores, and its Relation to Chemotaxis. Discussion on the Toxicity of the H-ions and Molecules of Acids.

In the chemotactic experiments described above, it has come frequently under my observation that the spores approaching the supraoptimal concentration of the attracting substances—those at the inner margin of the ring in the ring-collection and at the inner end of the column in the column-collection—had shrunken bodies, became less active and finally motionless followed by apparent death after a short length of time. This fact evidently points out that the spores have been driven to a diffusion-zone containing such amount of chemicals as is at once repellent and toxic to them, the amount being just above the optimum for positive chemotaxis. On the basis of this view I thought it possible to determine the exact liminal intensity of the concentration of the attractive agents given above as exerting repulsion, if the critical concentration of the same agents, which gives a similar effect upon the spores as is observed at the repulsion-

space, is in any way successfully found out. The present experiments will perhaps satisfy our aim on this subject.

When the active swarm-spores of *Aethalium* liberated in water are submitted to the action of several acids or other agents, they will exhibit various reactions according to their various concentrations. In order to make the given agents act uniformly on the whole surface of their body, the solution of these agents is added quickly, drop by drop, to the culture-medium of the spore, until the medium contains the required concentration of these agents. At a certain strong concentration we observe that the spores come immediately to rest, round up their body, become granular, and disorganize after a certain interval of time. If the solution is more dilute, the spores do not come to rest so rapidly: they first round up their body, move the cilia for a while, and then gradually come to rest, further changes then set in. When that medium is diluted before they reach the final stage, they soon resume motion. If the solution is still more dilute, the changes proceed more slowly; they become first dull in movement, their body deforming meanwhile, and further changes set in more slowly, until the final state is arrived at after 40-60 minutes. Something exactly similar is observable at the outermost repulsion-zone in chemotactic experiments, and hence, in the present case, our aim is to determine the critical concentration of the given agents. The following table gives the results of the experiments carried out with this view under the same condition, in particular under the same temperature (*ca.* 20°C.). For comparison there are given, besides chemotactic agents, other agents not immediately concerned, either indifferent or strongly poisonous.

Table IX.

Substance.	Basic value.	Concentration preventing the movement and causing contraction of the spores after 40-60 minutes.	Remarks.
Boric acid	3	1/10-1/15 mol	In 1/16 the spores are active till the next morning.
Malic acid	2	1/500	Many spores become motionless without deformation of body.
Succinic acid	2	1/500	
Tartaric acid	2	1/500	Many spores become motionless without deformation of body,
Hydrochloric acid	1	1/600	
Nitric acid	1	1/600	
Chromic acid	2	1/600	
Oxalic acid... ..	2	1/600	
Acetic acid... ..	1	1/600-1/700	
Phosphoric acid... ..	3	1/600-1/700	
Formic acid	1	1/600-1/700	
Lactic acid	1	1/600-1/700	
Citric acid	3	1/700	
Sulphuric acid	1	1/700-1/800	In 1/800 a few spores are active till the next morning.
Picric acid	1	1/900	
Salicylic acid	1	1/1500	In 1/1200 the spores perished at once but in 1/1600 they are indifferent.
Acid sodium sulphate ...		1/200-1/250	In 1/160 the spores are killed immediately; 1/300 is indifferent.
Monopotassium phosphate..		1/20-1/30	
Potassium bichromate ...		1/40-1/50	
Potassium nitrate		1/15-1/20	In 1/20 some spores may be alive till the next day.
Sodium acetate		1/20	In 1/25 the spores are alive till the next morning.
Sodium salicylate		1/20	In 1/30 the spores are active after 2 hours and 1/15 immediately kill them.
Cane-sugar		1/4-1/5	In 1/6 all alive; in 1/3 the body shrinks immediately.
Copper sulphate... ..		1/1600	The body rounds up.
Mercuric chloride		1/2600	The body soon rounds up.

Thus we see that of the several acids tested boric acid has the weakest and salicylic acid the strongest action in checking the free movement of the spores. The other acids do not stand very far apart from one another in this respect ranging from 1/500-1/900 mol.

From these results it is easily seen that the acid which has the strong concentration for checking the movement, like malic, tartaric, and succinic acids, allow the spores, in the experiment with the capillary tube, to advance nearer to the diffusion-centre than one of the weak concentration like sulphuric, picric, acetic acid, etc., the consequence being that the repulsion-space is smaller in the former. This assertion will with certainty confirm the results of experiments concerning the relative size of the ring, or in strict sense, of the repulsion-space, which has been mentioned above (Table V). Thus, of inorganic acids we have observed that sulphuric acid gives a larger ring than nitric, hydrochloric, and phosphoric acids, which is in coincidence with the fact that the former acid repels the spores at 1/700-1/800 mol, while the latter acids check the movement at 1/600 mol. Again, of organic acids the ring given by malic and tartaric acids is smaller than that given by lactic and formic acids. This difference in the size of the ring strictly accords with the result of the last experiment that the outermost repulsion-zone in the former acids is at 1/500 mol or thereabout, while it lies in the latter farther from the centre of the stimulus and is at 1/600-1/700 mol.

In this experiment it will become very evident that the repulsion-space given by oxalic acid is mainly, if not exclusively, due to the H-ions; for, its critical solution is equal to that of hydrochloric and nitric acids, and does not differ widely from that of acetic acid. Now, while oxalic acid is easily dissociable, acetic acid dissociates with difficulty, so that in equimolecular solutions of 1/600 or 1/700 mol we can not ascribe the repulsion in oxalic acid to the molecules remaining undissociated.

The repulsive concentration in formic and lactic acids is the same as in acetic acid (Table IX). Yet we observed that the repulsion-space in the former is smaller than in the latter (Table V). This is in contradiction to our expectation and seems to be owing to the circumstance

that in the former the larger amount of the H-ions attracts the spores so strongly as to drive them into the repulsion-space, where they soon come to rest and accumulate there, diminishing the apparent size of the repulsion-space.

Citric acid checks the movement at $1/700$ mol, and the ring is consequently nearly equal in size as in acetic acid. But in the former the H-ions are more numerous and more spores are collected in a given interval of time into a thicker and denser ring than in the latter.

It must be remarked that the shrinking of the spores entering the repulsion-space is not due to osmotic action, nor is the repulsion due to osmotaxis. The evidence for this statement may easily be found in the effects of concentrated solutions of some agents, such as some neutral salts or undissociable substances, which are indifferent in chemotaxis and non-poisonous. As examples of such agents we have given in the above table (Table IX) potassium nitrate, sodium acetate, sodium salicylate, and cane-sugar. They do not check the movement of or kill the spores except in considerably stronger concentrations, thus the three salts just mentioned check the movement at $1/15$ - $1/20$ mol and cane-sugar at $1/4$ - $1/5$ mol,¹ which are far from being isotonic with the critical solutions of the acids. At the given concentrations, for instance, of potassium nitrate and cane-sugar, their inhibiting action on the movement of the spores is in my opinion certainly due to the strong osmotic action rather than to chemical action. This confirms the view already set forth that the repulsion-space caused by the less dissociable acids is due not to the osmotic action but to the existence of the free molecules, and the same space caused by easily dissociable acids to the supraoptimal amount of the H-ions.

It is noteworthy that there is a remarkable difference in the manner of reaction of the spores to the repellent concentration of easily dissociable and less dissociable acids. While in the former acids the spores can escape

1. Cane-sugar seems to penetrate the plasmic membrane of the spores; they recover gradually from inactivity and move as before.

from the injurious action of supraoptimal concentration, in the latter they can not, as a rule, easily escape from it, so that they are mostly injured and come to rest sooner or later. The difficulty of the spores in getting out of the injurious zone of the latter acids seems to be due to the simultaneous action of the H-ions and the free molecules. In all probability the injurious effect must be attributed to the circumstance that the attraction of the H-ions overpowers the repulsion of the free molecules, so that the spores are driven into the fatal zone. A somewhat similar case was reported by BULLER ('00, p. 555) in his chemotactic experiment with fern-spermatozoids. He thinks that the repulsion from the acid substance is undoubtedly chemotactic and the repellent effect produced by the concentrated solution of the attractive salts may be osmotactic, and that the repulsion from the former substance is protective, while it is not in the latter substances, since the spermatozoids can not escape their injurious action. If the same explanation for the repulsion applies to the present case, we may say, reciprocally to BULLER's statement, that the repulsion from the concentrated solution of the H-ions is protective, while the repulsion from the free molecules of organic acids is less protective or perhaps not protective.

Let us now turn upon the consideration of the toxic action of acids. The components present in their solutions in the repulsion-space is surely toxic upon the spores, since they are killed by their action sooner or later, either by the H-ions or free molecules. According to the results obtained by a great many authors, the toxic activity of acids accords well, as PFEFFER noted ('04, p. 351), with their dissociability, with few exceptions, for instance, that of hydrocyanic acid whose anion may exert a specific toxicity. It means that the H-ions are the principal component concerned in the toxicity of acids¹. However, from several accounts which I have given above, I do not hesitate in saying that the toxic action of acids on the swarm-spores of Myxomycetes behaves differently accord-

1. The literature on this subject is enumerated and fully criticized by PFEFFER ('04, p. 339) and CZAPEK ('05, p. 902).

ing as the acids are strong or weak. Having found no parallelism of toxicity and dissociation at all, we must now first of all consider that the free molecules of weak acids may come into play in their toxicity. The toxicity of the molecules of some acids is not unknown. HEALD ('96, p. 138) has already expressed the view that the toxic effect of acetic acid is due to the undissociated molecules. CLARK ('99, p. 401) has shown that undissociated molecules of several acids are more poisonous than the acid ions. As to the relative toxicity of the molecules and other components in acid solutions there are in fact great variations in different organisms. Thus HEALD found that the molecules of acetic acid are less toxic than those of mineral acids (H-ions) on *Zea* and *Pisum*, confirming the results of KAHLENBERG and TRUE ('96) on *Lupinus*, while CLARK working on mould fungi came to the contrary conclusion that the molecules are more toxic than mineral acids. As to the relative toxic value of the H-ions and the molecules of acetic acid in *Myxomycetes*, definite result can easily be obtained by calculation. As the least toxic concentration of hydrochloric acid is 1/600 mol (Table IX), there must be contained an amount of the H-ions of about 0.167/100 gm. per litre. Now 1/700 mol of acetic acid, which is the least toxic concentration according to my experiments, will contain the undissociated molecules in amount a little more than 0.128/100 mol or less¹. It may be concluded, therefore, that the given molecules are more toxic than the H-ions. It must be remembered in drawing this conclusion that the presence of anions has no bearing upon it.

It is evident from the results of the experiments already described that the toxic value of the molecules of various acids is not all equal. Although 1/600-1/700 mol of lactic, formic, and acetic acids is the least toxic concentration, the amount of molecules at this concentration is smaller in the two former than in the last, as inferred from their dissociability. It follows that the molecules of lactic and formic acids are more toxic.

1. See OSTWALD ('89, p. 174.)

Boric acid does not show toxic action in the strict sense, since its 1/15 mol is the fatal strength for the spores. As its 1/16 mol is nearly isotonic with 1/20 mol potassium nitrate, the arrest of the motion of the spores must be due to the withdrawal of water from their body rather than to the chemical peculiarity of its molecules, the chief component in solution of such a weak acid. This fact confirms the view that boric acid is less poisonous than other acids (KAHLENBERG and TRUE, p. 107, and see also COPELAND and KAHLENBERG, '99, p. 470-471, CZAPEK, p. 918), or we may say that it is not poisonous in the sense we have considered.

The highly toxic effect of picric and salicylic acids will recall our special attention. Both acids being less soluble in water, I was not able to study the ring-collection, especially the relative size of the ring as compared with that given by other acids. Still it is possible from the results last mentioned to believe that they can produce larger repulsion-space, as we have found that their more dilute solution is powerful enough to bring about an arrest of motion, picric acid at 1/900 mol and salicylic acid at 1/1500 mol. The component concerned in repulsion can not be the H-ions; for though they dissociate comparatively easily, the amount of the H-ions evolved at equimolecular solution must be far smaller than in the typical mineral acids. Hence the remaining components, viz. free molecules or anions, must play the principal rôle in this respect. According to OSTWALD ('85, p. 354) picric acid dissociates almost completely at 1/1024 mol, pointing out that it is a strong acid and can attract as strongly as hydrochloric, nitric etc. It follows then that at 1/900 mol of this acid the undissociated molecules are present only in negligible amount, and consequently the anions are the principal toxic agent, the toxicity being stronger than that of the H-ions. In favour of this view it may be stated that in chemotactic experiments with this acid the spores approaching nearest to the centre of the stimulus immediately perished, showing beyond doubt the presence of strongly toxic components. The above view is moreover strengthened by the fact that the sodium salt of picric acid is also highly toxic, though the toxicity does not seem to be equal to that of the free acid,

being in fact somewhat less. As regards to toxic action of picric acid, therefore, I agree with TRUE and HUNKEL ('98), who conjointly worked on the toxic effect of picric acid and its sodium salt upon *Lupinus*, and came to the conclusion that the anions have very strong toxic effects (p. 392-393).

The strong toxicity of salicylic acid presents some peculiar features. In my experiments 1/1500 mol is sufficiently toxic upon the spores. While according to OSTWALD ('89, p. 247) this acid dissociates at 1/1024 mol 62.80 per cent., the critical solution given above dissociates more easily than 1/1024 mol. Compared, however, with the solution of hydrochloric acid 1/600 mol, the given solution must contain far less H-ions (*ca.* 1/4th.). Therefore, the toxic effect of salicylic acid at the given concentration must be due to the anions or free molecules. When KAHLLENBERG and TRUE's result that the toxic action of salicylic acid (p. 119) upon higher plants is of the same value as that of several inorganic acids, is taken into account, it appears that the H-ions are the essential toxic agent. But CLARK concluded that sodium salicylate is more toxic on mould than hydrochloric acid (p. 396), while TRUE and HUNKEL arrived at the conclusion that the sodium salt is less toxic than the acid, the critical strength for *Lupinus* being L/6400 for the acid and 1/100 mol for its salt (p. 394-395). If CLARK's result be correct, we must conclude that the anions or molecules are the active agents, but according to TRUE and HUNKEL's results it is probable that the anions do not have strong toxic effect. While these contradictory views are being entertained regarding the toxic component of salicylic acid, my experiments show that the relative toxicity of the acid and its salt is nearly as found by TRUE and HUNKEL, and that the undissociated molecules are the principal component concerned.

The boundary concentration of sodium salicylate is 1/20 mol, isomolecular with potassium nitrate, and bringing about an arrest of motion by withdrawal of water from the body of the swarm-spores. If the acid anions of this acid are strongly toxic, the same component that must be evolved in its salt should exert the same effect. The cases

being however otherwise, no other conclusion can be drawn than that the undissociated molecules of the acid are the toxic agents.

The inhibitory effect on the motion of the spores is very characteristic in malic, succinic, and tartaric acids. The other acids cause an immediate shrinking of the body before the motion is arrested, but in these acids the shrinking of the body is not the first effect on the spores. In the experiments on ring-collection we have observed that the spores at the inner margin of the ring become motionless without changing the form of their body, though some shrunken spores were found in the more internal region. It is certain, therefore, that the molecules of these acids urge the spores to continue their motion. This phenomenon is in strict agreement with the chemokinesis observed by ROTHERT ('01) in the swarm-spores of *Saprolegnia* reacting to meat-extract (p. 374). The motionless spores are not injured, so that they can easily resume active motion if they can free themselves in 30-40 minutes from the action of the concentrated solution of the acids. In ROTHERT'S experiments it is not apparent whether one and the same component of the meat-extract exercises both chemotactic and chemokinetic stimuli or whether there are two components each acting differently. In my opinion, in the case of Myxomycetes the two actions are exercised by different components, chemotaxis by the H-ions and chemokinesis by the free molecules. The molecules are, first of all, the cause of the formation of the repulsion-space by their chemokinetic action. It is, however, a matter of course that the higher concentration of the molecules may be toxic and kill the spores like other acids.

GARREY (p. 314) reported that *Chilomonas* is chemokinetic to almost all acids, while it is chemotactic to some of them (acetic, lactic, and butyric acids). He ascribed the former action to the H-ions, and the latter to the anions. As regards the active components for these different stimuli, I came to a different conclusion, viz. that the H-ions are concerned in tactic and the molecules in kinetic reaction. When we think that the chemokinetic phenomenon of *Chilomonas* was wholly denied by JENNINGS and MOORE ('02) who found that the H-ions act undoubtedly

chemotactically, instead of chemokinetically, we may inquire whether chemokinesis in *Chilomonas*, if present, is not related to the molecules as in *Myxomycetes*.

In this place I shall consider for a while the behaviour of the spores towards acidic salts. At several places I have stated that their attractive power is nearly the same as that of the free acids, but their repellent action is less remarkable in the equimolecular solutions. This is shown clearly in the last table. The boundary concentration for arrest of motion is $1/20$ - $1/30$ mol for monopotassium phosphate, and $1/50$ mol for potassium bichromate. In determining the toxic component of the latter salt comparison with chromic acid seems to be needed. KAHLLENBERG and TRUE (p. 106) have first noticed that chromic acid has the same toxic value upon higher plants as hydrochloric acid. As was mentioned in the table, this relation is also true in the case of *Myxomycetes*. Since chromic acid is weak, we can not ascribe the given toxic value to the H-ions. As to whether the anions or the free molecules are the toxic components, KAHLLENBERG and TRUE did not express any view. CLARK found that potassium bichromate and potassium chromate are nearly as toxic as mercuric chloride. He says, "As poisons for the mould they rank, as already mentioned, with formaldehyde, silver, and mercury" (p. 390). From this fact he concluded that the anions are very toxic; "The anion of the bichromate, Cr_2O_7 has a toxic value of about 770 H" (p. 390), thus agreeing with STEVENS' results of the study of the same salt as to its toxic effect upon fungi. My experiments lead me to results contrary to his that potassium bichromate is far less toxic than chromic acid, the boundary concentration being $1/50$ mol for the salt and $1/600$ mol for the acid. The salt is in highly dissociable and we may deduce that its anion is not toxic and for same reason we conclude that the high toxicity of chromic acid is due to the undissociated molecules. In my opinion, the stronger injurious effect of chromic acid as compared

with potassium bichromate upon *Paramecium* as ascertained by JENNINGS ('99 b, p. 359) may likewise be due to the same component.¹

Acid sodium and potassium sulphates kill the spores at a much weaker concentration than the salts given above, viz. at 1/200-1/250 mol. These salts are strongly acid in reaction and the toxicity may probably be due to the supraoptimal amount of the H-ions.

XI. Minimal Concentration for Chemotactic Stimulus.

As I have already discussed, phobotactic organisms are not sensitive to the direction of the diffusion of the stimulant and can not orientate themselves towards the source of the stimulant. This fact has led us to conclude that, to bring the spores to the mouth of the capillary tube, it is first of all necessary to form diffusion-zones, which cause frequent reversals of motion, irrespective of the amount of the stimulant at the mouth of and in the tube. In my opinion, therefore, usual capillary method is not available for the determination of the liminal value ("Schwellenwert") for the stimulus. This is justified by the results of the experiments shown in Table X, which were made by the method just referred to. The experiments were undertaken with *Aethalium* under a nearly constant temperature (20°C.). III denotes the column-collection in the tube; II, diffuse entry; I, slight collection in the tube; and O, no attraction or indifferent condition.

1. JENNINGS found that *Paramecium* is killed by 1% of potassium bichromate and 1/150 % of chromic acid in one minute.

Table X.

Acid.	Basic value.	Concentration of acids in the tube in mol.						
		1/100	1/200	1/300	1/400	1/500	1/600	1/800
Sulphuric acid	2		III		II		I	0
Citric acid	3		III		II		I	0
Malic acid	2	III	III-II		II-I	0	0	0
Succinic acid	2		III-II	II	II-I		0	0
Tartaric acid	2		III	II	I	0	0	0
Phosphoric acid	3	III		II-I	I-0	0	0	0
Chromic acid	2		III	II-I	0			
Picric acid	1	III	III-II	II-I	0			
Oxalic acid... ..	2		II	0 (I)	0		0	
Acetic acid... ..	1	III-II	I (II)	0 (I)	0			
Salicylic acid	1	III	II	I				
Nitric acid	1	III	II-I	0 (I)	0			
Formic acid	1	III	II	0				
Hydrochloric acid	1	III	I (II)	0				
Lactic acid... ..	1	III	II					
Boric acid ¹	1	0	0	0				
Water (for control)		0	0	0				

In this experiment we are struck with the remarkable result that the liminal value obtained by this method is in fact evidently toxic. For instance, while 1/600 mol of hydrochloric acid is so strong as to be injurious to the spores (Table IX), its 1/300 mol contained in the tube do not exert any attraction. Moreover, as another evidence against the acceptance of this liminal value we find that it is not isohydric in all the acids thus far examined. We may consider here that the distribution of the H-ions at the diffusion-zones given by isohydric solutions of several

1. 1/2 mol is the weakest concentration in attracting the spores in the tube.

acids is in variable states, in one case favourable and in the other unfavourable in attracting the spores into the diffusion-centre. We have already learned in the ring-collection that the acids in the diffusion-zone *ca.* 2 mm. distant from the mouth of the tube containing 1 mol are at the concentration of nearly $1/500$ - $1/600$ mol or $1/600$ - $1/700$ mol, when examined after 20-40 minutes.¹ It was also mentioned in another place that $1/20$ mol malic acid in the tube gives a diffusion-zone nearly at $1/500$ mol after 10 minutes just in the front of the mouth. So that it seems probable that, if a dilute solution, for instance, $1/200$ mol hydrochloric acid, contained in the tube be assumed to diffuse out with approximately equal velocity as a concentrated one, we will get roughly $1/200 \times 1/20 = 1/4000$ mol at the zone immediately surrounding the mouth, or $1/200 \times 1/600 = 1/120000$ mol at the zone 2 mm. distant from the mouth of the tube. Of course, it must be remembered that the manner of distribution of the H-ions in the diffusion-zones of acids is in fact highly complicated: the amount of the H-ions to be found at a certain zone may depend upon the valency of acid radicals, whether mono-, di-, or tribasic, the solubility of acids, in other words, affinity of acid molecules to water, and further the velocity of diffusion. Yet, from the calculations given above, we may say with some reason that the tube containing $1/300$ mol hydrochloric acid is unable to form round the mouth a diffusion-zone with sufficient amount of the H-ions to stimulate the spores to reverse their motion.² PFEFFER seemed to have been contented to find the "Schwellenwert" in both phobo- and topotaxis from the concentration to be found in the tube, though he pronounced the necessity to find it exactly in the diffusion-zones, which is however practically impossible ('84, p. 380). However, while at present we are aware of a great difference in the reaction to stimulus in phobo- and topotactic organisms and I convinced myself in my experiments already set forth that a most apparent inconsistency would

1. Compare Tables V and IX.

2. PFEFFER ('88, p. 588) has already noticed the exceedingly rapid decrease of the concentration of solutions at the mouth of the tube.

result, were the results obtained by the capillary method taken as valid, we come inevitably to the conclusion that the liminal value obtained by the latter method can not be accepted as an approximate value, since it is far above the boundary solution for toxicity. In consequence, we must now attempt to estimate the value with greater accuracy by a different method, which would give results more consistent with other physiological actions of the same agent.

It is more than evident that, in the determination of the liminal value in phobotactic organisms, the concentration at the outermost diffusion-zone, where the first turning of the stimulated organism occurs, must be taken as the least value for chemotactic stimulus, on which the concentration in the tube has in no way a direct effect. We can not yet by all means determine this critical concentration, but we will satisfy ourselves in accepting the following method which may give, at least in my case, more accurate result for our purpose than by the usual capillary method.

A comparatively wide tube (0.3-0.4 mm. in diameter) is immersed with one open end in the culture-medium, in which the active swarm-spores of *Aethalium* are densely swimming about; a large number of the spores is thus caught in the tube by the capillary action. After sealing one end of the tube the other end is inserted in several weak solutions of the acids to be tested. The acids outside the tube will enter it by diffusion, and the successive diffusion-zones will be regularly graded from the mouth inwards. We observe the reaction of some spores in the tube lying near the mouth to the decrease of the amount of the H-ions, as expressed by their turning movement. The spores that react in this manner are invariably those that are swimming in the tube away from the mouth inwards, that is, those which are leaving the field with a strong solution of the agent for one with a weaker solution. When we follow these spores in their forward motion, we will find that they stop suddenly after traversing a short distance, reacting to the decrease of the amount of the H-ions; they come to rest for a while, then resume their motion in some other direction, and finally swim towards the mouth of

the tube. The first turning shows that there is such a decrease of the H-ions as is sufficient to stimulate them. It must be noted that the first turning occurs at a point in the tube farther from the mouth, if the concentration of the agent outside the tube be increased. The distance between this point and the mouth is not same with different concentrations of the agent. However, it is certain that the concentration at that point is always constant, expressing the liminal value for positive phototaxis. The determination of that concentration is highly difficult, when the point lies in the tube. But if we regulate the concentration of the agent outside the tube so as to cause the first retreating movement at or near the mouth, we may assume that the spores feel the decrease of the concentration as soon as they proceed a short distance into the tube, and we may take this concentration as the nearest liminal value obtainable by the present method. The concentration of several acids thus determined is given as follows:

Sulphuric acid	1/20 000 mol.
Hydrochloric acid	1/10 000 mol.
Tartaric acid	1/8000-1/10 000 mol.
Malic acid	1/4000-1/6000 mol.
Acetic acid.. .. .	1/1000 mol.

The above result is strictly consistent with the dissociation theory; the necessary concentration of the acids is nearly isohydric.

To elucidate the details of the reaction of the spores when they are under the action of different concentration of an acid, I will note in the following an instance with hydrochloric acid. The experiment was made under a constant temperature (20°C.).

Concentration outside the tube.	Manner of reaction.
1/200 mol.	The spores gather densely in the tube 2 mm. deep from the mouth in 20-30 minutes.
1/400 mol.	The spores gather similarly 1 mm. deep after the same length of time.
1/600 mol.	The spores are collected similarly 0.5 mm. deep in the same length of time.
1/800 mol.	In 10 minutes a slight collection may be observed at the mouth.
1/2000 mol.	Turning occurs near the mouth; the collection is obscure, as the spores mostly get out of the tube and swim away.
1/5000 mol.	Nearly the same effect.
1/10 000 mol.	Turning is immediately observed near or at the mouth.
1/50 000 mol.	No reaction.
Distilled water (for control).	No reaction.

Thus 1/50000 mol hydrochloric acid is too weak to give in its diffusion-zones such differential distribution of the H-ions as the spores can respond to. To get the desired distribution the medium outside the tube is required to contain at least 1/10000 mol of the acid.

XII. Negative Chemotaxis with Alkaline Substances.

It has been already shown that the repellent action of alkaline substances is evidently due to the presence of the OH-ions in solution. As PFEFFER noted ('88, p. 598), the repellent action is generally less apparent than the attracting action. This was especially striking when very weak solutions were tested; it was sometimes not easy to determine

whether the spores were indifferent towards, or weakly repelled by that solution.

For this reason the method for determining the liminal value of the stimulus designed for positive chemotaxis is not applicable here. If, however, the liminal value for toxicity were determined, it would be clear beyond all doubt that the liminal value for the negative stimulus should be below that limit, and therefore, it is first of all necessary to determine the relative toxic value of alkaline substances as compared with acids.

With this view an attempt was made to ascertain at what degree of concentration of the given substances the spores become incapable in movement, just as we have seen in the toxic concentration of acids. Putting first the spores in $1/2000$ - $1/550$ mol of sodium hydroxide we observe that the body shrinks after 10-20 minutes, though they are still in motion. If a large amount of water is added to that medium, the spores recover soon. Next in $1/4000$ - $1/6400$ mol similar phenomena are observable. There are found, however, in $1/6400$ mol a few spores remaining unaffected and apparently healthy. The healthy ones have increased in number the next day, and after 2-3 days the most of the spores have recovered, giving an impression that they have become accommodated to the agent. Nearly same condition of the spores is observed in $1/4000$ mol, but in $1/3000$ mol only 50% recover.

In this connection it must be remarked that, in culture-medium containing alkaline substances, carbon dioxide may possibly accumulate owing to the respiration of the spores, in consequence of which the number of the OH-ions present in the medium become less and less. This is theoretically possible, though I am not able to measure the amount of the acid produced, and it gives ground for believing that the recovery of the injured spores in alkaline substances after a certain length of time is due to a decrease in the amount of the OH-ions caused by the neutralising action of the carbon dioxide.

At any rate, observed at the beginning of the experiment, we may say that the spores would escape from the region with $1/6400$

mol, since this solution evidently acts unfavourable to the motion of the spores; and as it is probable that repulsion must take place somewhat below the injurious concentration, it may be assumed that the spores would escape uninjured from the solution of about $1/10000$ mol.

We have shown that $1/10000$ mol hydrochloric acid is the liminal value for attraction and an innocuous concentration of that acid may extend below $1/600$ mol. It follows, therefore, that while $1/10000$ mol is the liminal value of attraction for hydrochloric acid, the same concentration is approximately maximal for sodium hydroxide, being repellent but not injurious. Hydrochloric acid may attract the spores without injury in the diffusion-zone slightly below $1/600$ mol. Hence it is not wholly impossible to draw from these the conclusion that the spores react negative-chemotactically to sodium hydroxide even if the latter is far weaker than $1/10000$ mol, so that they seem to be more sensitive to the OH-ions than to the H-ions.

Again, we may note in the present case the parallel effects of the toxic action of the two ions. PFEFFER ('04, p. 351) has already shown that their toxic actions are different upon different plants: according to CLARK the OH-ions are more toxic on mould than the H-ions, and *vice versa* on Bacteria and Phanerogams according to KAHLENBERG and TRUE. In Myxomycetes we have observed that $1/2000$ mol sodium hydroxide kills the spores in a few minutes, while $1/800$ mol hydrochloric or nitric acid does not, showing evidently that the OH-ions are here more toxic than the H-ions. This result, therefore, agrees with that of CLARK (p. 400) on mould.

XIII. Toxic Action of the Heavy Metals.

In the foregoing pages I have considered chiefly the toxic effect of chemicals concerned in chemotaxis. In comparison with them the study of some heavy metals is of special interest. As a rule, the heavy metals are known, as ascertained by many authors, to be much more toxic than

acids or other agents. We may ask then how they behave towards the swarm-spores of Myxomycetes as regards their toxic effect? In answering this question I shall give here the toxic effect of copper sulphate and mercuric chloride upon the spores of *Aethalium*.

In this experiment it is at once clear that these salts are strongly repellent even in very dilute solutions; they cause shrinkage of the spores or kill them immediately. 1/26000 mol mercuric chloride is strong enough to kill them immediately. However, its 1/100000 mol does not kill at once, inducing only shrinkage of their body. It is certain that such a dilute solution seems to repel the spores, since we can not find any spore entering that zone or moving with shrunken body. Copper sulphate causes shrinkage of the body at 1/16000 mol, the motion continuing however. Its stronger solution, of course, kills the spores sooner or later.

In this experiment we have ascertained that the toxic actions of the Cu- and Hg-ions are not equal, the latter being much more toxic.

As a whole the relative toxic values of H-, Cu-, and Hg-ions towards Myxomycetes are similarly related to one another as towards Phanerogams and fungi (KAHLENBERG and TRUE, HEALD, STEVENS, CLARK, etc.).

XIV. General Conclusions and Summary.

The intimate relation of acids to Myxomycetes recalls our attention to their biological importance for the existence of that organism. In the natural condition almost all species belonging to Myxogasteres develop on rotten or decaying plant-tissues, in the majority of cases on rotten wood. In the Botanic Garden at Koishikawa, Tokyo, where I have collected numerous species of Myxomycetes, I have always found them on wood which had been partly decomposed by the attack mostly of the higher fungi. Extracting a piece of this wood with boiling water I obtained a brownish acidic fluid. Again, on the wood where I found a very large clump of the sporangia of *Aethalium septicum* which furnished the chief material of the experiments described in the present article, I could find

in the autumn many large fructifications of a certain species of Polyporaceae. After saturating a block of this wood with water and pressing it, I extracted a dark brownish fluid, which, tested after boiling for some time, showed the presense of an acidic substance corresponding in amount to 1/150 normal solution of sodium hydroxide. The fluid showed a very strong attraction for the spores, evidently owing to the H-ions present. On the basis of such fact I can now point out two important actions of the H-ions as favourable for the development of the swarm-spores of Myxomycetes. In the first place the H-ions promote the germination of the spores and in the second place the swarm-spores thus liberated are attracted to the place where more H-ions are accumulating. While it is probable that the production of the H-ions is due to the decomposition of the wood and indicates the liberation of some organic matters supplying food-materials to Myxomycetes, it would be naturally conceived that the chemotactic sensitiveness of the swarm-spores to the H-ions should be an important factor in enabling them to reach the proper habitat, of equal importance, considered from the biological point of view, as the property in virtue of which many plant-spermatozoids are attracted by such substances as may be contained in the archegonium, or some fungus-mycelia bend towards certain nutritive substances.

The chief results of the present investigation may be recapitulated as follows:

1. The swarm-spores of Myxomycetes are attracted by all acids and acidic substances, and repelled by all alkalies and alkaline substances, while they react almost indifferently to all neutral substances, if they are not poisonous, the strength of the solution being assumed to be moderate in all cases.
2. The H- and OH-ions are the stimulating components in the substances experimented with.
3. The spores are more sensitive to the OH-ions. They are repelled by nearly 1/10000 mol, or perhaps below that, of sodium hydroxide, while the same concentration of hydrochloric acid is considered as the liminal value for attraction.

4. The H-ions contained in 1/600 mol of any well-dissociable acid are nearly the maximal amount for attraction. At higher concentrations they act repellently and injuriously.

5. The attracting power of acids is parallel to their dissociability, so that a strong acid has a stronger action than a weak one.

6. The repellent action of strong solutions of acids may be due in strong acids to the supraoptimal amount of the H-ions and in weak acids, in most cases, to undissociated acid-molecules.

7. The repellent power of acid-molecules is weaker than the attracting power of the H-ions, so that, when both components coexist in equal amounts, the spores attracted by the H-ions are injured by the coexisting acid-molecules.

8. The action of acid-molecules differs more or less in different acids; many of them act injuriously and cause shrinkage of the body of the spores at necessary concentrations, while others, for instance, malic, succinic, and tartaric acids, induce at first chemokinesis, that is, check of movement.

9. Many acidic salts (comprising such salts as give acidic reaction in solution by hydrolysis) are less toxic or repellent than those of free acids.

10. Both positive and negative chemotaxis is typically phobotactic, or to use JENNINGS' terminology, the reaction is a form of a "motor reflex."

11. In determining the liminal value ("Schwellenwert") of agents for chemotactic stimulus the usual capillary method of PFEFFER appears to be unreliable. The value found by this method is inconsistent with other physiological effects; for instance, it is larger than the toxic value, or it is not isohydric in all acids.

12. From the biological point of view the H-ions act beneficially to the development of Myxomycetes; they promote the germination of the spores on the one hand and lead the swarm-spores to the place where much food-materials are liberated on the other.

13. The toxic effects are different in different acids: the toxicity

is due either to the H-ions (many inorganic acids), undissociated molecules (many less dissociable organic acids), or anions (picric acid).

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June, 1909.

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THE HISTORY OF THE
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Studies on the Hybridology of Insects.

II. A SPORT OF THE SILK-WORM, BOMBYX MORI, L., AND ITS HEREDITARY BEHAVIOR.¹

BY

K. Toyama.

With Plate I.

Amongst our experienced silk-worm breeders it is known that certain breeds occasionally throw off some "red worms," but as to how they are produced and how they behaved in inheritance much obscurity still prevails.

In the year 1905 I found some red worms suddenly arisen from a cross between two normal black breeds, and thus a series of experiments was conducted with the intention of working out the laws of its inheritance. The results will be given in the following pages.

THE FIRST GENERATION.

(F₁).

In the spring of 1905 we crossed a Japanese univoltine breed called "*Datenishiki*" with the male of a tetravoltine breed, "*Tōbuhime*," and obtained 18 batches of eggs.

The newly hatched worms of both parent breeds are brownish black, i.e. normal in color. In the fifth stage, however, we have only commonly marked worms in the former breed, and both common and pale worms in the latter.

1. This paper is a continuation of Part I, which was published in the Bulletin of the College of Agriculture, Vol. VII, No. 2, 1906.

The eggs obtained from this cross hatched out on the 20th, 21st April of the next year (1906). All the worms when hatched were bluish black in color, that is to say, they remained true to the parents.

THE SECOND GENERATION.

(F₂).

The dark-worms derived from the cross above mentioned spun cocoons on the 23rd-26th May and the moths emerged on the 8th-10th June, 1906.

From these moths we obtained 627 batches of eggs which hatched on the 21st, 22nd June of the same year. The newly hatched worms of some batches, to my astonishment, were of two kinds, one quite normal in color and the other beautiful orange-red, without any intermediate forms.

The characteristics of the red worms (Pl. I. Fig. I): The head of the newly hatched worm is shining brownish red, the ground color of the body beautiful orange-red, the hair-bearing tubercles lighter than the ground color and the hairs or bristles light reddish-brown. After the first moult, the ground color of the body becomes whitish and in the later stages we find two kinds of worms, one having the common markings, and the other destitute of all markings, as in the pale worms of the common breeds. We can distinguish, however, full grown worms of the normal breeds from the red, since the markings which are dark brown in the former are light brownish-red in the latter (Figs. III and IV).

From some matings, we get a dark-red form. In this form the ground color of the body is brownish-red, while the other characters are quite the same as in the normal red form. This is a newly hatched worm of the red form which exhibits common markings in later larval stages.

In the common breed, however, the head of the newly hatched worm, as is represented in Fig. II, is shining black, the ground color of the body smoky black, the hair tubercles light brown and the hairs and bristles light smoky.

For the sake of convenience, we shall call hereafter the former as the "*red worms*" and the latter or normal worms the "*black worms*."

Of the 627 matings before mentioned some, e.g. No. 6, gave rise to two kinds of worms, red and black, while others produced black worms only, as is shown in Table I.

Table I.

No. of parentage.	No. of egg-batches from each parent.	Date of hatching.	Kind of worms hatched out.
1	134	21-22 June, '06.	Every batch produced simply black-worms.
2	139	do.	do.
4	102	do.	do.
7	115	do.	do.
6	137	do.	110 batches produced only black-worms, 27 batches red and black-worms in the proportion shown in Table II.

Each batch is the total of the eggs laid by a moth.

The number of the red and black-worms given by each mating of parentage No. 6 is given in Table II.

Table II.

No. of egg-batches.	Kind of worms hatched from each batch.		Total number of worms hatched out.
	Black worms.	Red worms.	
1	356	100 (21.92%)	456
2	383	108 (21.99%)	491
3	353	80 (18.47%)	433
4	358	92 (20.66%)	450
5	329	85 (20.53%)	414
6	268	95 (20.66%)	363
7	285	109 (27.66%)	394
8	346	103 (22.93%)	449

No. of egg-batches.	Kind of worms hatched from each batch.		Total number of worms hatched out.
	Black worms.	Red worms.	
9	296	85 (22.36%)	380
10	311	107 (25.59%)	418
11	314	101 (24.33%)	415
12	267	87 (24.57%)	354
13	225	63 (21.87%)	288
14	265	68 (20.42%)	333
15	241	79 (24.37%)	320
16	217	73 (25.17%)	290
17	240	73 (23.32%)	313
18	329	76 (18.74%)	405
19	327	97 (22.87%)	424
20	205	56 (21.45%)	261
21	346	102 (22.76%)	448
22	237	58 (19.66%)	295
23	} Both red and black worms were produced, but the number was not recorded.		
24			
27			
25	315	94 (22.98)	409
26	350	112 (24.24%)	462
Total.	7,162	2,103 (22.69%)	9,265
Expected Mendelian figures.	6,948	2,316	

These figures immediately suggested that we had to do with the phenomenon of Mendelism, with the black worm dominant over the red. The result of the next generation will confirm this conclusion.

The offspring from other matings which yielded simply black worms remained true to the parents in the subsequent generations and never once produced any red-worms, as far as my experiments went.

THE THIRD GENERATION.

(F₃).

Both red and black worms found in each mating of the second generation were reared separately. The mounting took place on the 16th-18th July and the moths emerged between 29th July and 2nd August 1906. 69 matings of the black worms and 65 matings of the red worms were made, of which the former laid the eggs which yielded uniform (Black only) and mixed (red and black) offsprings, and the latter only red worms, as is shown in Table III.

Table III.

Parentage.	Kind of parent worms.	Emergence of moths et oviposition.	Hatching of the eggs.	Kind of worms hatched out.
25	Black.	29/7-2/8/'06.	16-17/4/'07.	{ Of the 58 matings obtained from this parentage, 35 yielded only black worms, and the rest red and black worms. Of the 25 matings, all produced red worms.
	Red.	do.	do.	
26	Black.	do.	do.	{ Of the 11 matings, 9 yielded only black worms, and 2 red and black worms. Of the 13 pairings, 12 produced simply red worms, while only one red and black worms nearly in the proportion of 1 red : 1 black. It must be supposed, however, that this was due to accident.
	Red.	do.	do.	
Mixed parentage.	Red.	do.	do.	{ Of the 27 batches, all produced uniform red worms, no black ones. These are brown shaded red worms as described before, and bred true to parents in following generations.

The number of both red and black worms found in each black mating is shown in Table IV and V.

Table IV.

Number of red and black worms found in each pairing of parentage No. 25.

No. of egg-batches.	Number of red worms.	Number of black worms.	Total.
8	53 (25.6%)	154	207
9	53 (22.45%)	183	236
12	73 (20.11%)	290	363
15	60 (24.59%)	189	244
17	66 (24.81%)	200	266
6	70 (28%)	180	250
7	32 (27.11%)	86	118
8''	54 (24.1%)	170	224
11	79 (23.93%)	251	330
14*	2	401	403
15 ¹	90 (24.45%)	278	368
16	98 (24.62%)	300	398
19	66 (22.93%)	309	375
20	82 (27.51%)	216	298
23	78 (22.28%)	272	350
25*	18	225	243
27	26 (20%)	104	130
29	65 (32.27%)	137	202
33	27 (16.16%)	140	167
39	43 (25.14%)	128	171
41	83 (22.49%)	286	369
46	44 (11.42%)	341	385
Total,**	1,242 (22.78%)	4,214	5,456
Expected number from the Mendelian theory.	1,364	4,092	5,456
Discrepancy.	- 122	+ 122	

** Excluding those marked with an asterisk.

Table V.

Number of red and black worms in each black mating of parentage No. 26.

No. of egg-batches.	Number of red worms.	Number of black worms.	Total.
9	54 (20.45%)	210	264
10	67 (24.18%)	210	277
Total.	121 (22.36%)	420	541
Mendelian expectation.	435	406	541
Discrepancy.	- 14	+ 14	

From the results of these two generations, I think, it is sufficiently clear that the "black" is dominant over the "red" and that the segregation of both characters in the offspring follows MENDEL'S law of heredity.

It is important to notice, however, that sometimes there occurred an incomplete segregation of the two characters. In Nos. 14 and 25 described in Table IV the segregation of the recessive character is not complete, as was the case in No. 29 for the dominant character. Similar cases of seeming exception to MENDEL'S law are mentioned by CORRENS.¹

Of the 65 pairings of red worms, all yielded uniform red offspring, except one mating No. 20 from parentage No. 26, which produced 187 reds and 194 blacks, that is to say, nearly in the proportion of 1 red : 1 black. I accounted it to be a case $(D+R) \times R$ which is due to the mixing of some hybrid black worms to the red group during the rearing. If so, every mating of the black worm should produce red and black worms in the proportion of 1 red to 3 blacks in the next generation. The result of the next generation will confirm this.

¹ Ber. d. Deutsch. Botan. Gesellsch., Bd. XX, 1902.

THE FOURTH GENERATION.

(F₄)

In this generation, we kept for experiment

- I. Red worms from parentage No. 26 of the last generation.
- II. Red worms from the mixed parentage of the last generation.
- III. { a. Red worms, { from the mating No. 2 of parentage No.
 b. Black worms, { 26 Red worm, which produced red and
 black worms in nearly equal proportion
 (see Table III).

They mounted on the first to fifth, Sept. 1906 and the moths emerged 18-22nd Sept. 1906.

72 pairings were made, in which the offspring of the red matings remained constant, while those from the black ones produced, as we expected, red and black worms in the proportion of 3 blacks to one red. Table VI and VII give the details of the experiment.

Table VI.

Offspring from the mating between red worms I and II.

No.	Date of oviposition.	Number of egg-batches.	Date of hatching.	Kind of worms hatched out.
I.	14/9/'06.	3	16/4/'07.	All red worms, no black.
	18-22/9/'06.	5	do.	do.
II.	17/9/'06.	32	24/4/'07.	do.

40 matings produced red worms with no exception. From the first appearance they remained true to the parent.

Table VII.

Worms from egg-batch No. 2 of parentage No. 26, which produced red and black worms in the proportion 1R: 1B.

a. Mating between red worms.

Date of oviposition.	Number of egg-batches.	Date of hatching.	Kind of worms hatched out.
18-13/9/'06.	16	7/10/'06. et 17/4/'07.	all red worms.

They also remained true to the parents.

b. Mating between black worms.

No. of egg-batches.	Date of oviposition.	Date of hatching.	Kind of worms hatched out.		Total.
			red worms.	black worms.	
1	18-23/9/'06.	7/10/'06. et 24/4/'07.	134 (28.68%)	334	468
2*	do.	do.	33	128	161 (the rest eggs died.)
3	do.	do.	64 (29.35%)	154	218
4*	do.	do.	11	40	51 (the rest died.)
5	do.	do.	85 (27.97%)	226	311
6	do.	do.	70 (28%)	180	250
7	do.	do.	81 (25.87%)	232	313
8	} do.	do.	203 (25%)	622	830
9					
10					
11*	do.	do.	36	125	161 (the rest died.)
12	do.	do.	5	31	36 (this is the total.)
13	do.	do.	63 (22.5%)	217	280
14	do.	do.	102 (26.7%)	280	382
15*	do.	do.	39	41	80 (the rest died.)
16	do.	do.	27 (23.4%)	88	115
Total. ¹			839	2,364	3,203
Mendelian expectation.			800	2,402	
Deviation.			+ 38	- 38	

1. Excluding those marked with an asterisk.

Now we see that every mating produced red and black worms nearly in the proportion of one red to three blacks and thus the supposition in the pervious generation was proved to be true.

BACK-CROSSING.

I. Hybrid black \times red,

or

DR \times R.

♀ Dark worm from III of the fourth generation.

♂ Red worm from I of the fourth generation.

Their results are given in Table VIII.

Table VIII.

No.	Oviposition.	Hatching.	Kind of worms hatched out.		Total.
			red worms.	black worms.	
1	17/9/'06.	7-8/10/'06.	190	198	388
2	do.	do.	165	146	311
3	do.	do.	125	137	262
Total			480	481	961

As the Mendelian principle demands, every mating gave red and black worms in the proportion of 1 Red to 1 black.

II. Hybrid black \times pure black,

or

D \times RD.

a. ♀ Red worms from I of the last generation.

♀ Pure black worms (a cross between Tetravoltine race and Theophila Mandarin).

b. ♀ Pure black worms, as above mentioned.

♂ Red worms, as above mentioned.

The results are given in Table IX.

Table IX.

No. of mating.	Oviposition.	Hatching.	Kind of worms hatched out.		
♀ ♂ Red Black	1	16/9/'06.	24/4/'07.	Simply black worms, no red ones.	
	2	15/9/'06.	do.	do.	
	3	do.	8-9/10/'06. et 24/4/'07.	do.	
	4	16/9/'06.	24/4/'07.	do.	
	5	do.	do.	do.	
	6	17/9/'06.	do.	do.	
	7	do.	do.	do.	
	8	do.	do.	do.	
	9	do.	do.	do.	
	10	do.	do.	do.	
	11	30/7/'06.	23/4/'07.	do.	
	13	17/9/'06.	do.	do.	
	14	do.	do.	do.	
	15	do.	do.	do.	
	17	30/7/'06.	do.	do.	
	18	do.	do.	do.	
	♀ ♂ Black Red.	12	do.	do.	do.
		16	do.	do.	do.

Every mating yielded only black worms. They mounted on 21-22nd May, 1907 and the moth emerged 6-12th, June, 1907. From these moths 231 matings were obtained. All the matings produced red and black which yielded only black worms.

The respective number of red and black worms found in each mating is given in the following tables:

Table X.

No. of mating.	Date of oviposition.	Date of hatching.	Number of worms hatched out.		Total.
			Red worms.	Black worms.	
1. G.II. 2	8/6/'07.	22-23/6/'07.	78 (26.89%)	212	290
2. 11	do.	do.	100 (23.76%)	285	385
3. 12	do.	do.	85 (29.82%)	200	285
4. 17	do.	do.	90 (26.70%)	247	337
5. 10	do.	do.	96 (25.26%)	284	380
6. 7	do.	do.	99 (26.82%)	270	369
7. GKII. 1	do.	do.	44 (24.58%)	135	179
8. 3	do.	do.	97 (28.00%)	249	346
9. 4	do.	do.	99 (25.78%)	285	384
10. 6	do.	do.	101 (24.10%)	318	419
11. GTh.II. 1	do.	do.	114 (28.21%)	290	404
12. 7	do.	do.	45 (20.04%)	179	224
13. 3	do.	do.	102 (33.00%)	207	309
14. 9	do.	do.	51 (23.18%)	169	220
15. 4	do.	do.	89 (25.35%)	262	351
16. 10	do.	do.	56 (34.78%)	105	161
17. X	do.	do.	81 (27.45%)	214	295
18. 9	do.	do.	80 (23.52%)	260	340
19. 13	do.	do.	35 (25.36%)	103	138
20. 10	do.	do.	52 (26.00%)	148	200
21. 22	do.	do.	80 (23.39%)	262	342
Mendelian expectation.			1,674 (26.32%)	4,684	6,358
			1,589	4,768	

Table XI.

No. of matings.	Date of oviposition.	Date of hatching.	Number of worms hatched out.		Total.	
			Red worms.	Black worms.		
1. G.I.D. 9	9/6/'07.	22-24/6/'07.	102 (26.35%)	285	387	No. of unfertilized eggs.
2. 2	do.	do.	77 (23.19%)	255	332	
3. G.I. 1	do.	do.	83 (24.05%)	262	345	
4. 9	do.	do.	99 (24.75%)	301	400	
5. G.I.D. 1	do.	do.	59 (22.51%)	203	262	
6. 3	do.	do.	88 (25.07%)	263	351	111.
7. 4	do.	do.	76 (23.10%)	253	329	111.
8. G.I. 10*	do.	do.	0	all blacks.		
9. 11	do.	do.	66 (27.60%)	173	239	180.
10. 12	do.	do.	63 (19.26%)	264	327	
11. 13	do.	do.	103 (24.93%)	310	413	
12. 14	do.	do.	103 (22.10%)	382	485	
13. 15	do.	do.	62 (22.54%)	213	275	
14. 16	do.	do.	127 (30.38%)	291	418	
15. 17	do.	do.	120 (27.64%)	314	434	
16. 18	do.	do.	95 (26.91%)	258	353	
17. 8	do.	do.	76 (22.75%)	258	334	
18. G.I.D. 5	do.	do.	87 (22.12%)	246	333	
19. 22	do.	do.	96 (22.91%)	323	419	
20. 23	do.	do.	86 (25.00%)	258	344	
21. 24	do.	do.	108 (24.65%)	330	438	
22. G.I. 22	do.	do.	54 (22.97%)	181	235	
23. 24	do.	do.	77 (23.69%)	248	325	
24. 25	do.	do.	105 (23.50%)	340	445	
25. 26	do.	do.	77 (19.79%)	312	389	
26. 27	do.	do.	92 (29.11%)	224	316	
Total.			2,181	6,747	8,928	
Mendelian expectation.			(24.42%) 2,232	6,696		

* The single mating which produced only black offspring must be supposed to be due to accident.

Table XII.

No. of mating.	Date of oviposition.	Date of hatching.	Kind of worms hatched out.		Total.
			Red worms.	Black worms.	
1. EIII. 3	8/6/07.	22-23/6/07.	121 (27.43%)	320	441
2. 4	do.	do.	113 (26.96%)	306	419
3. 19	do.	do.	97 (25.66%)	281	378
4. 21	do.	do.	112 (23.52%)	362	474
5. 24	do.	do.	94 (23.26%)	310	404
6. EIII.a. 1	do.	do.	56 (21.87%)	200	256
7. 2	do.	do.	114 (27.60%)	299	413
8. 3	do.	do.	109 (26.91%)	296	405
9. 4	do.	do.	99 (25.31%)	292	391
10. 5	do.	do.	105 (26.51%)	291	396
11. 6*	do.	do.	1	345	346
12. 7	do.	do.	88 (27.84%)	228	316
13. 8	do.	do.	123 (26.91%)	334	457
14. 9	do.	do.	74 (20.68%)	187	261
15. 10	do.	do.	86 (22.99%)	288	374
16. 11	do.	do.	82 (25.15%)	244	326
17. 12	do.	do.	109 (24.66%)	333	442
18. 13	do.	do.	91 (23.94%)	289	380
19. 14	do.	do.	103 (24.23%)	322	425
20. 15*	do.	do.	168 (37.52%)	277	445
21. 16	do.	do.	90 (24.72%)	274	364
22. 20	do.	do.	110 (28.20%)	280	390
Total. ¹			2,144 (26.29%)	6,013	8,157
Mendelian expectation.			2,039	6,117	

In Nos. 6 and 15, we again see instances of incomplete segregation of the antagonistic characters.

1. Excluding that marked with an asterisk.

Table XIII.

No. of mating.	Date of oviposition.	Date of hatching.	Kind of worms hatched out.		Total.
			Red worms.	Black worms.	
1. EI. 1	9/6/'07.	23-25/6/'07.	11 (25.92%)	321	432
2. 4	do.	do.	88 (23.30%)	289	377
3. 12	do.	do.	100 (26.30%)	280	380
4. 11	do.	do.	95 (23.00%)	318	413
5. 10	do.	do.	101 (29.50%)	265	366
6. 9	do.	do.	108 (25.10%)	319	427
7. 8	do.	do.	93 (22.79%)	315	408
8. 7	do.	do.	102 (26.42%)	284	386
9. 6	do.	do.	81 (24.03%)	256	337
10. 5	do.	do.	87 (26.28%)	244	331
11. 15	do.	do.	82 (20.39%)	320	402
12. 16	do.	do.	88 (23.34%)	289	377
13. 17	do.	do.	86 (33.33%)	172	258
14. 18	do.	do.	97 (24.80%)	294	391
15. 19	do.	do.	105 (25.42%)	308	413
16. 20	do.	do.	103 (24.40%)	319	422
17. 21	do.	do.	99 (26.40%)	276	375
18. 22	10/6/'07.	24-26/6/'07.	100 (26.59%)	276	376
19. 23	do.	do.	108 (24.05%)	341	449
20. 24	do.	do.	82 (20.39%)	320	402
21. 25	do.	do.	88 (23.34%)	289	377
22. 26	do.	do.	82 (22.22%)	287	369
23. 27	do.	do.	88 (24.44%)	272	360
Total.			2,174 (24.62%)	6,654	8,828
Mendelian expectation.			2,207	6,621	

Total worms derived from four parentages GI, GII, EI and EIII:

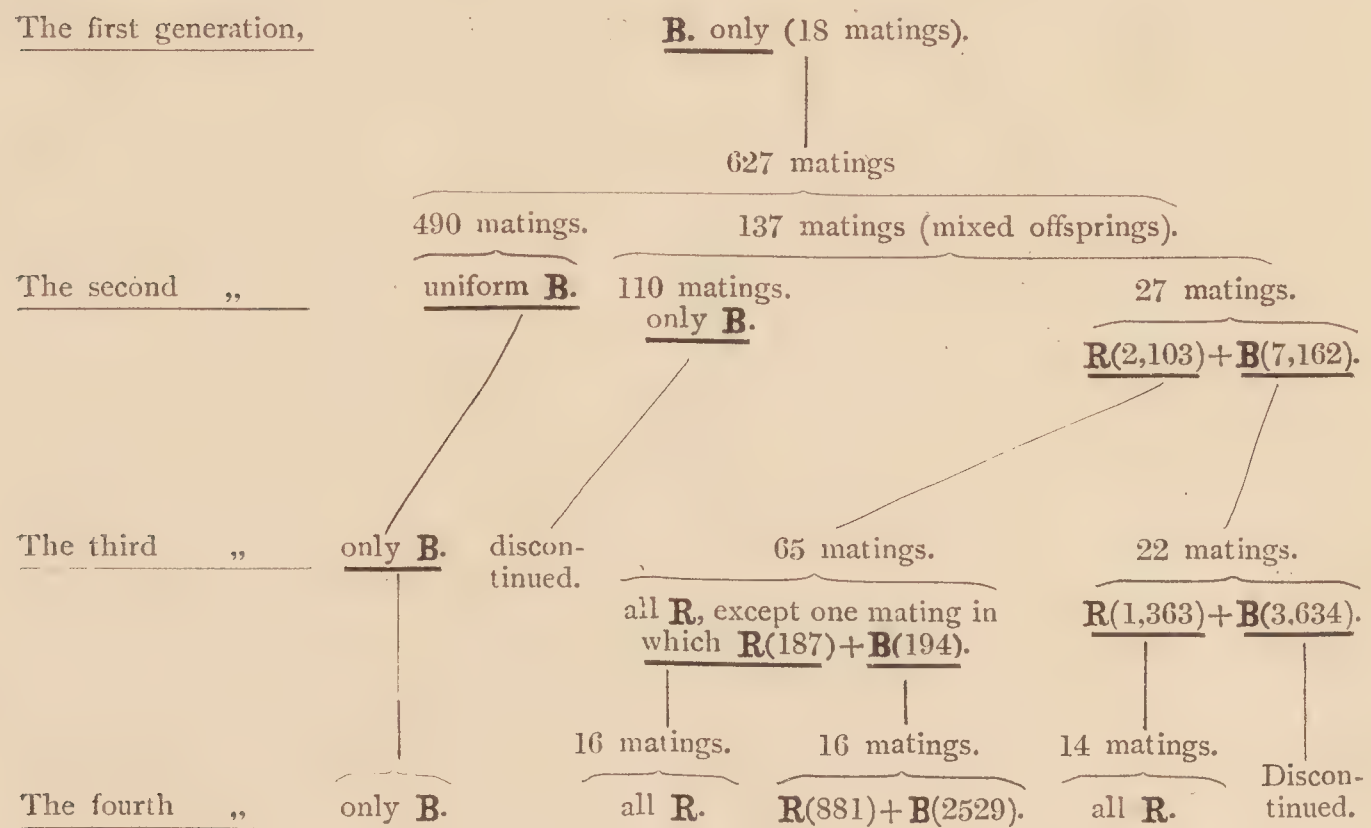
			Red worms.	Black worms.	Total.
GI.	(Table IX)	25 matings	2,181	6,747	8,928
II.	(Table X)	21 „	1,674	4,684	6,358
EI.	(Table XI)	23 „	2,174	6,654	8,828
EIII.	(Table XII)	22 „	2,144	6,013	8,157
Grand Total.....			8,173	24,098	32,371
Mendelian expectation			8,067	24,203	
Deviation			+105	—105	

Now we see clearly that the character “red” is recessive towards “black,” a character of different lineage, according to MENDEL’S law of heredity.

Résumé of the results obtained on the inheritance of the characters, red and black worms:

♀ UNIVOLTINE BLACK WORMS × ♂ TETRAVOLTINE BLACK WORMS.

B=Black worms; **R**=Red worms.



BACK-CROSSING.

(1) ♀ Hybrid blacks × ♂ reds.

Offspring reds(480) + blacks(481).

(2) ♀ Pure blacks × ♂ reds.

(Reciprocal crosses.)

The first generation, All blacks (15 matings).

231 matings

The second „ reds (8,173) + blacks (24,098).

Summary.

1. The sport "red worms" may be arisen from black breeds by crossing which apparently bring about the segregation of the dormant character.

2. The red worm thus produced remains constant from its first appearance. It is recessive toward the ordinary black worms and segregates from the latter according to the Mendelian law of heredity.

3. Sometimes it happens that the segregation of both characters is not complete, as in the case already observed by CORRENS¹ in Plants.

4. In the experiment described in a previous paper² we secured a new form quite different from the parents. We have now again procured red worms. We may mention another case in which two Mendelian recessive characteristics, "blue" and "brindled" (*Kasuri*) worms arose spontaneously from the normal worms. All of them, however, were result of crossing and are quite constant from their first appearance. Any instance of pure mutation not yet observed. Hence we are now inclined to believe that the sports which are commonly met with in the silk-worm mostly is due to hybridization, that is to say, hybrid mutation as LIDFORSS³ says, and not mutation in the sense of DE VRIES.

1. CORRENS, Scheinbare Ausnahmen &c. Ber. Deutsch Botan. Gesellsch., Bd. XX, 1902.

2. The first part of this contribution. (Bull. of Coll. of Agric. Vol. VII, No. 2, 1906.)

3. B. LIDFORSS, Studier öfver artbildningen &c. 1905.

Zoological institute,

College of Agriculture,

Tokyo Imperial University.

Explanation of Plate.

Plate I.

Fig. 1. Represents a young larva of the red form in the first stage (2 days old). Magnified about eight times.

Fig. 2. Represents a young dark larva of the ordinary form in the first stage (two days old). Magnified about eight times.

Fig. 3. Full grown worm of the red form. Markings on the body are decidedly reddish brown. Nat. size.

Fig. 4. Full grown worm of the black form. Markings are blackish brown. Nat. size.

Vol. II. No. 1, PUBLISHED DECEMBER 8th, 1909.

Price in Tokyo Yen 0.60.

THIS JOURNAL IS ON SALE AT

Z. P. Maruya & Co., L'td.

Tori Sanchoe, Nihonbashi, Tokyo.

明治四十二年十二月六日印刷
明治四十二年十二月八日發行

編纂兼發行者 東京帝國大學

印刷者

西谷 弟 松

東京市麻布區市兵衛町二丁目十六番地

印刷所

シヤパン、タイムス社

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13,851

Vol. II. No. 2.

東京帝國大學
農科大學紀要

第二卷第二號

JOURNAL

(FORMERLY BULLETIN)

OF THE

COLLEGE OF AGRICULTURE,

IMPERIAL UNIVERSITY OF TOKYO.

TOKYO.

Published by the University.

FEBRUARY 12TH, MEIJI XLIII (1910).

PUBLISHING COMMITTEE.

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On the Pathology of the Jaundice (Gelbsucht) of the Silkworm.

BY

Prof. **C. Sasaki**, *Rigakuhakushi*

With Plate II—VII.

Introduction.

The silkworm disease known as Jaundice (Gelbsucht), prevails in all the countries where the worms are bred, and in our country it is very frequently met with, being of annual occurrence. It is called in Japanese "Nobyō," and the diseased worms are known as "Nosan" or "Umiko."¹

It is very characteristic that the blood of the diseased worms becomes turbid—in the yellow or green races, the turbid blood bears more or less a yellowish, and in the white race, a whitish color. Several authors have worked on the pathology of this disease, but their opinions still differ on several points.

My studies on this subject were begun in 1893 in conjunction with my friend, director J. BOLLE of Görz, who visited Japan in the same year. They were continued for some years, but I could not devote all my time to the subject on account of my work on other subjects. Since 1903, I have again taken up the subject and spent some time upon the pathology of the disease.

Before entering into the details of my studies it will not be entirely useless to refer to the results so far obtained both by Europeans and Japanese, so far as my knowledge goes.

1. "Umiko": Umi means pus, ko silkworm.

In 1808, Mr. P. N. NYSTEN, who seems to have been one of the earliest observers on this disease described its symptoms as follows:—

“La maladie à laquelle on donne particulièrement le nom de Jaunisse, s’observe constamment dans toutes le magnauderies, vers la fin du cinquième âge, c’est-à-dire, lors que les vers, pleins de la matière soyeuse, se disposent à filer. Cette maladie consiste dans une teinte jaune et une bouffissure de tout le corps du ver, c’est une espèce d’anasarque ou une infiltration du liquide untritif dans toutes les parties de l’animal. Elle commence autour des stigmate, se propage de là aux articulations des anneaux, que se relivent en bourrelets en jaunissant, et bientôt elle gagne toutes les parties du corps. Les pieds paraissent alors raccourcis à cause du gonflement des parties environnantes, et le ver jaune n’exerce qu’avec peine tous ses mouvements.....”

In 1825, Count DANDOLO³ says “There are other disorders, called Giallume riccione, and the Soffocamento; the two first are modifications of the above-described, and only differ in violence, or by the circumstances which produced them; however, in these diseases, the silky substance is seldom effected.....”

The symptoms of the Jaundice given in L’ABBE ROZIER’S *Cour d’Agriculture*, which are extracted by the translator of DANDOLO’S work⁴, are as follows:—

“Of the Vaches, Gras, or Yellows: 1st. The head of the worm swells. 2nd. The skin is drawn tight over the rings, and shines as with varnish. 3rd. The rings swell. 4th. The circumference of the aperture of the stigmates is of light or deep yellow colour. 5th. And the worm voids a yellow liquid which may be seen on the leaves.”

Mr. CARLO-PICHIAT⁵ wrote on the symptoms of Jaundice thus:—
“Soltanto allorché inferiscono l’altre malattie, non si presente il giallume: nel quale la tumefazione, nidi il color giallo, infine lo scoppiar delle

2. P. H. NYSTEN: *Recherches sur les Maladies de Vers à Soie* 1808.

3. Count DANDOLO: *The Art of rearing Silkworms* (English Translation). p. 272. 1825.

4. Ditto. p. 278.

5. R. LAMBRUSCHINI: *Il Baco filo*. p. 209. 1853.

pelle vesando un liquor giallo, e fetido, costituiscono il processo morboso ond'è tratto a perire. Dubito che questo liquido giallo sià lo stesso umor serico alterato, perchè ne'bigatti da seta bianca, il color giallo è infatti biancastro." The opinions of Mr. E. CORNALIA⁶ are as follows:—"La trama fibronosa che costituisce i globuli li echinati, di cui parlai all'articolo de sangue, si sciolgono e lasciano in libertà i granuli che contengono, de quali s'aggruppano nel medesimo tempo che scompajono tutti gli altri elementi che rinvengonsi nel sangue del baco nell stato di sanità. Lo stato chimico del sangue affetto del giallume continue ad essere piuttosto acido fino alla totale sua decomposizione, esso è opaca, lattiginosa simile a tenue pus. Oramai quasi tutti i baco-fili conven-gono che la impedita traspirazione e la impedita ossidazione dei principii combustibili dell'atto respiratorio siano le cause del giallume; dipendenti poi queste o dalle circostanze atmosferiche viziate, o da una muta imperfetta della pelle, certamente le cause principali, sotto le quale pare si sviluppi di preferenza il giallume, sono da un lato l'aria tumida, gli ambienti non ventilati, la troppo poca luce, la poca pulizia; dall'altro, l'azione della muta, e specialmente della quarta, che dà il segnale dello sviluppo del giallume."

Prof. F. HABERLANDT⁷ described the symptoms of the disease as follows:—"Bei solchen fettsüchtigen Raupen (Vacce) beginnt die Krankheit mit einer Verdickung der Vorderringe, welche sich bald über den ganzen Körper erstreckt, die Haut verdickt sich gleichfalls, wird opak, verfärbt sich bei Gelbspinnern in weissliches gelb, das indessen den Körper nie so gleichmässig färbt, als bei der früher geschilderten höhergradigen Erkrankung. Das Blut ist auch bei den specksüchtigen Raupen milchig getrübt....."

In 1872, Prof. HABERLANDT⁸ published the results of his studies on the Jaundice. He says, "Zu den sporadisch vorkommenden Krankheiten, welche in der Regel eine Minderzahl der Raupen befallen und

6. E. CORNALIA: Monographia del Bombyce de Gelso. p. 348. 1856.

7. F. HABERLANDT: Der Seidenspinner p. 238. 1871.

8. Oesterreichische Seidenbau-Zeitung 1. April 1872.

nur in seltenen Fällen und unter besonders günstigen Umständen ganze Aufzuchten dahinraffen, gehört die *Gelbsucht*.... Auch die Auflösung des Fettgewebes der gelbsüchtigen Raupen, welche wir früher vermuthet hatten, indem wir von der Voraussetzung ausgegangen waren, dass die Trübung des Blutes von einer Bemischung zahlreicher aus dem Fettkörper ausgetretener Fetttröpfchen herrühre, ist eine unbegründete gewesen, indem die milchige Beschaffenheit des ausfliessenden Blutes vielmehr von einer grossen Anzahl kleiner, dem Blute beigemischter Krystalle herrührt." Further Prof. HABERLANDT adds the followings:—"Schon CORNALIA sah diese Krystalle wie aus folgender Beschreibung des Blutes gelbsüchtigen Raupen.....Er bemerkt dort, dass sich bei Gelb- oder fettsüchtigen Raupen insbesondere das Blut verändere. Die stacheligen Blutkörperchen sagt er, lösen sich auf und lassen die Körnchen frei, welche sie enthalten.... Diese Körnchen, welche CORNALIA auf Tafel XV in Fig. 265 ganz getreu abbildet, sind eben jene mikroskopischen Krystalle,.... Dr. VERNON hat zuerst die Krystallnatur dieser Körnchen vorkommen erkannt*." In 1875, Mr. J. BOLLE⁹ described the Jaundice thus:—"Die von der Gelbsucht befallene Raupe verliert allmählig ihren Appetit und verschmäht schliesslich jegliche Nahrung. Ihr Körper, anstatt wegen mangelnder Nahrung an Umfang abzunehmen, schwillt immer mehr und mehr an, bis die Haut, da sie eine weitere Ausdehnung nicht mehr erlaubt, platzt und dem Leibe eine Flüssigkeit entquillt, die wenn die Raupe zu einer Race gehört, die gelbe Cocons liefert, gelb, weiss gefärbt aber dann ist, wenn die Raupe den Weiss-oder Grünspinnern angehört. Nach dem Tode wird die Raupe schwarz, die inneren Organe derselben zerreißen und verwandeln sich in eine stinkende schwarze Jauche. Die Grundursache dieser Krankheit kennt man noch nicht, doch stimmen alle Seidenzüchter darin überein, dass eine mangelhafte Ventilation, niedere Temperatur und allzu grosse Feuchtigkeit der die Raupen umgebenden Luft die Entwicklung der Gelbsucht sehr begünstigen....."

* The crystal nature of the Körnchen is later discussed by R. PANEBIANCO in Bolletino mensile di Bachicoltura N. 11 1895, and also by the same author in Rivista di Min. e cristall. Italiana Vol. XXIX.

9. J. BOLLE: Die Krankheiten des Maulbeerseidenspinners. 1875.

Prof. E. VERNON¹⁰ characterised the Jaundice as follows:—"Il sangue ha perduto la naturale sua limpidezza, e si é convertito in quell'umore lattiginoso ch'esce dalle lacerazione della pella. E la causa di tale interbidamento sta nella presenza d'immuroveli corpicciuoli sospesi nel liquido, che generalmente furono ritenuti goccioline di grasso. All'incontro è risultato da nostre reserche, che le presunte goccioline di grasso sono minutissime formazioné crystalline di natura finora ignota..... Intorno alle causa del giallume regna assoluta oscurita....."

Dr. de FILIPPI¹¹ mentioned his observations on the same subject (Jaundice) thus:—"Voici une autre observation bien plus intéressante, dont le résultat est constant, et que chacun peut répéter facilement dans les vers atteints de la maladie appelée *Jaunisse*. Dans ces vers, le fluide circulant est réduit à une vraie émulsion, colorée en jaune vif par la quantité innombrable de globules de grassie qui s'y tiennent en suspension. Or ces globules, non seulement se rencontrent toujours et en abondance dans l'espace intermembranulaire, mais ils s'y montrent avant d'être dans le fluide circulant lui-même, et ils proviennent évidemment d'une production graisseuse des cellules peritoneals, ou bien, à ce qu'il semble, de leurs noyaux, car dans les vers atteints de cette maladie ces noyaux se montrent, comme dans la fig. 7....."

In 1881, director J. BOLLE¹² of Görz, wrote a brief account of the Jaundice as follows:—"Bei erkrankten Raupen, welche den Gelbspinner-Rassen angehören, färbt sich die Haut gelb, während bei den Raupen der Grün- oder Weissspinner den Körper undurchsichtig und weiss wird. Die Haut zeigt sich leicht zerreisslich und lässt sich trübe milchige Flüssigkeit ausfliessen....."

Mr. E. ROCHEBRAVE¹³ described a symptom of the Jaundice as follows:—"L'apparition de ces deux maladies est préparée par les mêmes

10. E. VERNON: Del Filugello e del suo Allevamento 1877.

11. Dr. DE FILIPPI: Insect en general et en particulier sur le Ver à Soie du Murier 1879.

12. J. BOLLE: Kurz Anleitung zur rationellen Aufzucht der Seidenraupe.

13. EMILE ROCHEBLAVE: Maladies des vers a soie 1893.

causes qui predisposent a la flacherie, avec cette différence que celle-ci est déterminée par le froid ou par un changement brusque du chaud au froid, tandis que les autres le sont par un changement brusque du froid au chaud excessif. La grasserie se déclare le plus souvent au 3^e âge. Les vers ont le corps blanc, boursoufflé; ils refusent de manger, de s'aliter, leur peau devient luisante, couleur verdâtre, ils transpirent une matière gluante et périssent. La Jaunisse arrive à la fin de l'un des âges, la peau quoique non relâchée complètement ne laisse apercevoir aucune ondulation sur le ver entre les anneaux de la tête a la queue. Le Sang cesse de circuler; de clair et limpide qu'il est à l'état sain il devient jaune trouble et d'un mauvais aspect; en quelques cas il devient noir, passant de la jaunisse a l'état de fondu."

Messrs. F. A. WACHTL and KARL KORNAUTH,¹⁴ after studying the so called Wipfelkrankheit of Nonne, has pointed out that this disease has a close similarity with the Jaundice which was studied by director J. BOLLE in 1873. The above mentioned two authors add further thus:—"Denn ist es nicht leicht möglich, an kranken Nonnenraupen stets eine Farbenveränderung wahrzunehmen, ferner scheint auch die Gelbsucht der Seidenraupen nicht mit dem Tod der Raupe zu enden, weil in dem Jahrbuche angegeben wird, dass mit Hilfe des Mikroskopes an dem Vorhandsein der polyedrischen Körnchen in den Puppen erkannt werden könne, ob dieselben von gelbsüchtigen oder schlaffsüchtigen Raupen herrühren."

Director J. BOLLE¹⁵ published again the result of his study in 1894 and concluded that the polyhedral body is a species of Coccidæ and a real cause of the Jaundice (Gelbsucht.) His observation on this body is as follows:—"Queste vescichette presentano tutti gli stadi intermedi di sviluppo dei granuli poliedrici contenendo esse cioè dapprima un liquido

14. F. A. WACHTL und KARL KORNAUTH: Beiträge zur Kenntniss der Morphologie, Biologie und Pathologie der Nonne (*Psilura monacha* L.) 1893.

15. JOHANN BOLLE: Jahrbuch der K. K. Seidenbau-Versuchsstation in Görz für das Jahr 1873. (I am very sorry this Jahrbuch is not accessible to me).

16. Atti e Memorie dell' I. R. Società agraria di Gorizia. Anno XXXIII. N. 5. 1894.

quasi salino, fornito di vacuole, poi pallide e minutissime granulazioni, quindi alcune di queste si fanno un po' più obscure, ingrandiscono e nello stesso tempo appajono goccioline sferiche, in gran numero, dapprima pallide poscia a contorno alquanto oscuro, tutte della stessa grandezza e riempianti l'intera suddetta vescichetta..... Infine si scorgono i granuli polyedrici, che sono la resultanza finale di questo sviluppo e di queta pressione, racchusi da una sottilissima membrana entro vescichette ovali."

"Lo sviluppo di questi granuli non è quindi altro che una regolare moltiplicazione, quale la si riscontra in molti infimi animali e precisamente della classe degli sporozoi e crediamo che corrisponda alla famiglia dei coccidi, in cui la moltiplicazione avviene in cisti, che sareberro le nostre vescichette, ed il cui indiduo adulto assume forma di spora, la quale nel nostro caso è il granulo poliedrico."

In 1895 I¹⁷ observed the polyhedral bodies in the fatty and muscular tissues, peri-tracheal membrane, silk glands &c. and I thought they were formed within the above mentioned organs as a result of insufficient respiration. My later studies¹⁸ on the Jaundice have taught me that the polyhedral bodies are produced within the internal organs of the silkworms as a result of certain diseases and unhealthy conditions, and that they are not of parasitic nature, but merely a sort of crystalloids.

Mr. J. M. KRASSILSCHTSCHIK,¹⁹ who found a microbe in the diseased silkworms, stated as follows:—"Le Sang des vers à soie malades de la grasserie présente assez souvent une culture pure d'un microcoque tout a fait minuscule (de 0'', 5 à 0'', 6 de diametre), que nous appellerons *Micrococcus lardarius*, et qui est le microbe specifique de cette maladie."

In 1898, director J. BOLLE of Görz, after a thorough study of the Jaundice during the preceding four years, has pointed out the parasitic nature of the polyhedral bodies (polyedrischen Körnchen).

17. C. SASAKI: Nosan (Jaundice) (Japanese) 1895.

18. Ditto: Nosan Kenkiu Yoho (Preliminary Report on the Study of Jaundice) (Japanese) 1905.

19. J. M. KRASSILSCHTSCHIK: Sur les Microbes de la Flacherie et de la Grasserie des Vers à Soie. Compt. Rend. p. 425—429. 1896.

From his work,²⁰ the followings are cited:—"Die braune Flüssigkeit im Leibe einer seit einigen Tagen an Gelbsucht verendeten Raupe zeigt, mit einem geringen Quantum destillirten Wassers verdünnt, unter dem Mikroskope bei 500-600-facher Vergrößerung ausser dem üblichen Detritus von Raupenleichen eine unzählige Menge kleinwinziger Körnchen, welche nach ihrer eigenthümlichen Gestalt von uns seit dem Jahr 1873 *polyedrische Körnchen* benannt worden sind, eine Benennung, welche nach uns von aller Fachmännern, die über die Gelbsucht der Seidenraupe geschrieben, einstimmig acceptirt wurde. Diese polyedrischen Körnchen sind ein constantes Symptom der genannten Krankheit und treten in gesunden oder mit anderen Krankheiten behafteten Raupen nie auf. Ihr Vorhandsein ist in allen Krankheitsstadien leicht erkenntlich, und nimmt ihre Zahl mit dem Vorschreiten der Seuche ungemein rasch zu. Auf sie ist die zunehmende charakteristische Trübung des Blutes von gelbsüchtigen Raupen zurückzuführen."

As a method of multiplication he says²¹ "Untersucht man Blutropfen von einer gelbsüchtigen Seidenraupe oder die Flüssigkeit einer Raupen— beziehungsweise Puppen-Leiche von einem an natürlicher oder künstlich eingepfelter Gelbsucht verendeten Seidenspinner, so entdeckt man unter der Unmasse von normalen polyedrischen Körnchen solche, welche wir ihrer eigenthümlichen Gestalt wegen Zwillingskörnchen heissen wollen.... Der Einschnitt erscheint dann schon wie echte Scheidewand mit dunkler Färbung, deren fast ovale Durchschnittslinie deutlich wahrzunehmen ist, wenn das durchsichtige Zwillingskörnchen, welches vordem einen mehr rundlichen Umriss hatte, eckiger.....und es zeigt sich dann, dass es thatsächlich aus zwei mit je einer ihrer Körperflächen anruhenden Einzelkörnchen besteht. Der Zusammenhang zwischen den beiden Einzelkörnchen lockert sich bei jenen Zwillingskörnchen, welche nahe daran sind, auseinanderzufallen (Fig. 45,e); mitunter gelang es uns den Abtrennungsmoment zu erschauen (Fig. 45, e)." The same author observed the second method of propagation of the polyhedral bodies and

20. J. BOLLE: Der Seidenbau in Japan. p. 105. 1898.

21. Loc. cit. p. 113.

stated as follows:—"Zweimal gelang es uns, die Aussonderung der protoplasmaartigen Masse unter der Form eines unregelmässigen, blassen, leicht körnigen kleinen Klümpchen direct wahrzunehmen; dasselbe trennte sich von Körnchen, als wäre es von diesem mit Gewalt hinausgeschneit worden, änderte seine Form, wir können nicht recht sagen, ob wegen seines kreisens in der Flüssigkeit oder wegen eines innern, sonst den Amoeben eigenthümlichen Bewegungstriebes. An der Absonderungsstelle des Amoebenformigen Klümpchens liess das Körnchen eine Oeffnung mit hervorragenden Rändern deutlich wahrnehmen....." Further he says²²:—"Das kernige Aussehen gewissen Tropfen ist so ausgeprägt, dass es dem Aussehen der mit typischem Protoplasma gefüllten Zellen gleichkommt; andere Tröpfchen unter den grösseren sind schon echte Bläschen mit doppeltem Umrisse, somit mit einer Hülle versehen und etwas glänzende kleine Kernlein, sowie zahllose verhältnissmässige grosse, schwach glänzende Kügelchen enthaltend.....Andere Bläschen sind endlich mit dunklen Körnchen von sechseckigen Umrissen besetzt, welche auch an Grösse mit den polyedrischen Körnchen identisch sind (Fig. 46, 1, m, n, o)."

As the 3rd method of reproduction, he says²³:—"Die Sporulae (Tröpfchen) verwandeln sich also in Cysten, ebenso wie die Amoeben von Sporozoaren einer höheren Ordnung....."

As a result of his studies, he²⁴ gave first the name *Microsporidium polyedricum* to these polyhedral bodies.

In 1903, director J. BOLLE²⁵ published some further studies on the parasitic nature of *Microsporidium polyedricum* and proved that this parasite had an infective power on several other insects such as *Bombyx*, *Autherea*, *Attacus* &c.

22. Loc. cit. p. 120—121.

23. Loc. cit. p. 128.

24. Loc. cit. p. 131.

25. Bericht über die Thätigkeit der K. K. Landw.-chemischen Versuchsstation. p. 8. 1903.

MESSRS C. MIYABARA and T. YANAI²⁶ (1903), Mr. J. OMORI²⁷ (1905), and Mr. K. TAMURA²⁸ (1905-1906), on the basis of their studies on the pathology of the Jaundice (Gelbsucht), have affirmed the disease to be due to the presence of a parasitic sporozoan. That is, they agree mostly with director J. BOLLE of Görz.

In 1907, Dr. S. PROWAZEK²⁹ proved that the Jaundice of the silkworm is due to the presence of *Chlamydozoan bombycis*. His studies on *Ch. bombycis* are abstracted as follows: "Am Rande dieser ausstrichpräparate aus der Leibeshöhlenflüssigkeit und dem Blut gelbsüchtiger Seidenraupen, wo das Serum in einer etwas dünneren Schicht fixiert worden ist, konnten, sobald die Präparate noch mit Giemsa's Eosinazur mehrmals hintereinander intensiv gefärbt und wie trockene Deckglas ausstriche weiter behandelt worden sind.....in dem Serumgerinnsel helle Gebilde in sehr grosser Zahl nachgewiesen werden (Fig. 2). In den hellen, ovalen bis runden, kleinen Gebilden konnte meist ein rotvioletter oder dunkelblauer punctartiger Körper von der Gestalt eines Coccus festgestellt werden. Diese Körper teilen sich zuweilen hantelförmig und ich sehe diese Gebilde als die *eigentlichen Erreger der Gelbsucht* der Seidenraupen an."

"Sie sind rundlich, vermehren sich durch eine hantelförmige Querteilung und scheinen eine gallertige Hülle (heller Saum) zu besitzen. Ich renne sie vorläufig *Chlamydozoan bombycis*."

MESSRS. A. CONTE et D. LEVRAT³⁰ have affirmed, after careful studies, that the polyhedral bodies (granules) characteristic of the Jaundice are the product of the degeneration of adipose and other tissues. Further they communicate thus:—"Nous retrouvons, dans chaque cellule, un

26. C. MIYABARA and T. YANAI: Studies on the Pathology of Jaundice (Japanese). Special Report of Fukushima Sericultural School. 1903.

27. J. OMORI: Brief account on the Pathology of Jaundice (Japanese). Report of Dai Nippon Sanshi Kwaiho. Nos. 42, 43, 44. 1905.

28. K. TAMURA: Studies on Jaundice. Report of Dai Nippon Sanshi Kwaiho. No. 178. 1907.

29. S. PROWAZEK: Chlamydozoa II. Gelbsucht der Seidenraupen. Archiv f. Protistenkunde, Band X. 2 u. 3. p. 363. 1907.

30. A. CONTE et D. LEVRAT: Les Maladies du Ver a soie. La Grasserie. Laboratoire d'Études de la Soie. Vol. XIII. p. 57-56. 1906-1907.

amas de crystalloïdes entouré d'une membrane qui se colore en bleu et, dans chaque amas, un petit corps de forme irrégulière prenant fortement la matière colorante. Ces réactions nous éclairent sur la signification de tels amas intraprotoplasmiques: ces sont les noyaux des cellules adipeuses; la membrane périphérique est la membrane nucléaire, le petit corps prenant fortement la matière colorante est formé de chromatine. C'est le reliquat de la substance nucléaires.....dans certaines de ces cellules (Pl. III), on voit d'abord des cellules intactes, puis des cellules à noyaux hypertrophiés, à substance chromatique agglomérée en amas irréguliers."

"Dans ces cellules, on voit apparaître de très petits points noirs, qui ne sont autres que les débuts de crystalloïds; c'est ce dernier stade que l'on voit à droite en haut de la planche. Puis, le nombre des petits crystalloïds augmente, leurs dimensions également. Il est à remarquer que les crystalloïds d'une même cellule sont à peu près tous de mêmes dimensions. Au fur et à mesure de la croissance des crystalloïdes, le noyau s'hypertrophie, la membrane se distend, et cela jusqu'à l'éclatement de la cellule, duquel résulte la chute du noyau dans le plasma sanguin."

As mentioned before, the Jaundice of the silkworm has been studied by several authors, but the cause or origin of the disease appears to be still unsettled, and I think it is still indispensable to spend much time in finding the true cause of this interesting subject. The results obtained by me on the subject are as follows:—

Symptoms of Jaundice.

As it is already well known among silkworm breeders, the worms affected by the Jaundice gradually lose their appetite, weaken gradually and finally die. At the end of the disease, the skin losing its firmness and becoming soft and weak, will be easily torn by slight touches, and as a result of this, a turbid blood flows out of the body. Thus the main symptom of the Jaundice is believed by almost all authors to be the

presence of a turbidity or milkiness in the otherwise clear blood, and of polyhedral bodies or crystalloids in the blood as well as in other organs and tissues; but according to my researches, the essential symptom of the disease is the presence of polyhedral bodies in the blood or other organs and tissues without reference to the quality of the blood. This point I will discuss later on.

The principal symptoms of the Jaundice, which are usually observed in our silkworms (white race) are as follows:—

1st: The diseased silkworms appear largely in the 5th age. Usually one or more but sometimes all the segments of the body are swollen up, and present more or less a milky white color. Finally, the skin, becoming weak and powerless, may easily tear off, and turbid white bloods flow out, and the worms will die.

The diseased worms as mentioned above are called by Japanese “Fushiko” or “Fushidaka.”

2nd: Most frequently the diseased worms may appear at each moult. In this case, the diseased worms are unable to go through the moulting process, and remain more or less of smaller size than the normal or moulted ones. The body is usually so swollen up as to make the segment lines hardly noticeable, besides being of a characteristically shiny white appearance. Sometimes the diseased worms are nearly the same size as the healthy ones, but we can easily distinguish the former by the smaller size of the head. If we cut open the body of the diseased worms, a milky white blood will flow out as an indication of Jaundice.

The worms showing the above mentioned symptom are called in Japanese “Nemuradzu” or “Hikaruko.”

The dead silkworms or pupae, which are imprisoned within their cocoons, contain often the polyhedral bodies (crystalloids), and further the parasitic maggots* that come out of the contained silkworms or pupae also contain the crystalloids within their body.

* These parasitic maggots make a great damage every year in Japan.

Nature of Polyedral Bodies (Crystalloids).

The polyhedral bodies or crystalloids which are found without exception in the silkworms or pupae affected by Jaundice are, as already described by Messrs. E. VERNON, J. BOLLE and others, flattened thin many sided plates; but they look mostly six sided. They are colorless, glossy and transparent, and brittle in nature (Fig. 1, Pl. II.) If we place a drop of the milky fluid (blood) of the diseased silkworms on a glass plate and strongly press the drop by means of a cover glass, then the polyhedral bodies may be easily crushed as represented in Fig. 2, Pl. II. If we dry up in the air the milky fluid on a glass plate, some of the crystalloids will show the same effect as mentioned above. Furthermore if we expose the crystalloids to the temperature of 150° C. for twenty hours or 140° C. for ten hours, most of the crystalloids are crushed or sometimes show various cracks or fissures. The crystalloids are heavier than water, viz., if we add a few drops of the milky blood to a clear water contained in a test tube, and shake it, the water will immediately become turbid; but if kept very quiet for some hours, the turbid water will again become transparent, leaving a white precipitate at the bottom of the tube. This precipitate consists of nothing else but the crystalloids.

The crystalloids are very variable in size. Some are extremely minute and granular, while the larger ones vary from 0.0066 mm. to 0.008 mm. in diameter.

They stain more or less with several reagents, having special affinities for carbol-fuchsin, picric acid, and their combination as double stains. The picric acid stains them usually yellow, while by the double staining of fuchsin and picric acid, the crystalloids stain red in the centre and yellowish in the peripheries.

Some say that the crystalloids are to be disintegrated by certain reagents; but according to my experiments, they are not affected by concentrated sulphuric or nitric acids, caustic potash (10%), saliva (of

man), ether, chloroform &c. Also they do not undergo any change by the application of alcohol, viz.: I took some diseased worms, which had been kept in alcohol (90%) for three years, and found that the crystalloids contained within their body retain still the normal shape.

Are the Crystalloids of the Silkworms, affected by Jaundice, of a Parasitic Nature?

It is admitted by almost all authors that the turbidity of the blood of diseased silkworms is the symptom of the Jaundice, and the granules are produced by dissolution or degeneration of the fatty tissues. The crystalloid nature of the granules was first announced by Prof. E. VERNON in 1872.

Director J. BOLLE made public the results of his studies in 1894, that the granules (*granuli poliedrici*) are a sort of Coccidae. Two years later, Mr. J. M. KRASSILSCHTSCHIK found a microbe, which he regarded as characteristic of the diseased worms, and he gave it the name of *Micrococcus lardarius*.

In 1898, director J. BOLLE gave to the granules (so called polyedrischen Körnchen) the name *Microsporidium polyedricum* from the consideration that the granules are the developmental stages of certain Sporozoa. Messrs. T. MIYABARA, T. YANAI, J. OMORI and others support in certain respects the view of director J. BOLLE of Görz.

Dr. S. PROWAZEK, having observed coccus-like bodies within the body of the diseased silkworms, has concluded the Jaundice of the silkworms is caused by a parasitic coccus, which he named "*Chlamydozoan bombycis*."

In 1907, Messrs. A. CONTE et D. LEVRAT mentioned the results of their studies on Jaundice in the Laboratoire d'Études de la Soie, viz:—The crystalloids of the diseased silkworms are produced within the nucleus of the adipose tissues and others as a result of hypertrophy.

My results on the study of Jaundice are entirely different from

the opinions of Messrs. J. BOLLE, S. PROWAZEK and others, according to which the Jaundice is caused by the presence of a parasitic protozoan; but agree in certain respects (but not all) with those of the non-parasitic nature of Jaundice.

In the following lines, I will describe the researches which I have done on the subject during the past years.

Are the polyhedral bodies in the diseased silkworms a sort of spores? To solve this question, I tried to see whether the polyhedral bodies germinate in the blood, juices of the stomach of silkworms or silkworm extract bouillon by sowing them in a hanging drop, but they remain always unchanged and never show an exit of protoplasmic mass, as director J. BOLLE has first observed.

On the Contagion of the Milky Fluid of Diseased Worms.

Experiment I. On the 24th May 1906, 20 healthy silkworms of Awobiki race (3rd day of the 4th age) were taken and the milky fluid taken from diseased worms* diluted with distilled water was injected under the skin of the silkworms.

The results obtained by this experiment† were as follows:—

Date.	Nos. of days in the 4th age.	Nos. of diseased worms.
24th May.	3rd day.	0
25th „	4th „	0
26th „	5th „	0
27th „	6th „	0
28th „	7th „	0
29th „	8th „	0
30th „	9th „	all affected by Jaundice.

Experiment II. On the 25th August 1906, 30 healthy silkworms (Awobiki race) on the 3rd day of the 4th age were taken and chopped

* "Diseased worms" always means those affected by Jaundice.

† The same experiment was repeated in August of the same year upon a quadrivoltin race (4th breed), and the same result was obtained.

leaves sprinkled over with the milky fluid diluted with distilled water, were given to them. The results were:

Date.	Nos. of days in the 4th age.	Nos. of diseased worms.
25th August.	1st day.	0
26th „	2nd „	0
27th „	3rd „	0
28th „	4th „	1 Jaundice.
29th „	5th „	„
30th „	6th „	0

On the 30th August, 20 worms mounted. On the 2nd September, 3 worms were found dead in the cocoon; all of them contained the crystalloids.

Experiment III. On the 15th May 1907, 30 healthy silkworms (Awobiki race) on the 1st day of the 4th age were taken, and the fresh milky fluid taken out of diseased worms was subcutaneously injected. The results were:

Date.	Nos. of days in the 4th age.	Nos. of diseased worms.
15th May.	1st day.	0
16th „	2nd „	0
17th „	3rd „	0
18th „	4th „	0
19th „	5th „	0
20th „	6th „	0
21st „	7th „	0
22nd „	8th „	0
23rd „	9th „	all diseased.

9 days after the injection, all the worms exhibited the characteristic symptom of the diseased silkworm, viz.: the body was swollen up and the contained blood was turbid and milky white. They lost appetite and commenced to crawl about out of the seat.

Experiment IV. On the 15th May 1907, 30 healthy silkworms (Awobiki race) on the 1st day of the 4th age were taken, fresh milky fluid of the diseased worms was sprinkled over the chopped leaves, and the latter were given to the worms. The results were:

Date.	Nos. of days in the 4th age.	Nos. of diseased worms.	Remarks.
15th May.	1st day.	0	
16th „	2nd „	0	
17th „	3rd „	0	
18th „	4th „	0	
20th „	5th „	0	
21st „	6th „	0	
22nd „	7th „	0	ended 5th moult.
23rd „	8th „	0	
24th „	9th „	0	
25th „	10th „	0	
26th „	11th „	0	
27th „	12th „	0	
28th „	13th „	2 diseased.	
29th „	14th „	4 diseased.	
30th „	15th „	0	24 worms mounted.

On the 7th June, of the 24 mounted silkworms 15 effected pupation, 9 died. Of the 9 dead worms 8 were infested by maggot and 1 contained crystalloids.

Experiment V. On the 17th May, 1907, 50 silkworms (Awobiki race) on the 3rd day of the 4th age were taken, and subcutaneous injection repeated exactly in the same way as in Experiment I. The results were:

Date.	Nos. of days in the 4th age.	Nos. of diseased worms.
17th May.	3rd day.	0
18th „	4th „	0
19th „	5th „	0
20th „	6th „	0
21st „	7th „	0
22nd „	8th „	0
23rd „	9th „	30 diseased.
24th „	10th „	20 diseased.

On the 7th and 8th days after the injection, all the silkworms showed the symptom characteristic of Jaundice, and after crawling out of the seat died finally.

Experiment VI. On the 17th May 1907, 50 silkworms (Awobiki race) on the 3rd day of the 4th age were taken, and fresh milky fluids were given to them with chopped leaves as in the Experiment IV. The results were:—

Date.	Nos. of days in the 4th age.	Nos. of diseased worms.	Remarks.
17th May.	3rd day.	0	
18th „	4th „	0	
19th „	5th „	0	
20th „	6th „	0	
21st „	7th „	0	
22nd „	8th „	0	ended 4th moult.
23rd „	9th „	0	
24th „	10th „	2	
25th „	11th „	0	
26th „	12th „	0	
27th „	13th „	5	
28th „	14th „	5	
29th „	15th „	1	
30th „	16th „	2	

During the experiment, 15 worms became diseased and the remaining 35 mounted, of which 20 effected pupation, 14 were infested by maggot, and 1 was lost.

By the above mentioned 6 experiments, it is proved that the milky fluid (blood) of the diseased worms has a strong contagious power, this being especially evident in experiments I and V. Similar experiments were carried on by director J. BOLLE³¹ in 1898, and he concluded that the contagion was due to the presence of crystalloids (polyedrischen Körnchen), which he considered as the cause of Jaundice.

It is still doubtful whether the crystalloids suspended in the milky blood of the diseased worms are to be looked upon as the cause of Jaundice, for it is very probable that the diseased worms may contain within their milky blood besides the crystalloids some other pathogenic micro-organisms, which cause the disease. In order to solve this question, I tried to isolate the crystalloids, but without success, but this

31. J. BOLLE: Der Seidenbau in Japan. p. 100. 1898.

attempt enabled us to isolate all the micro-organisms contained in the blood if such be present.

In fact, 4 species of microbes were obtained from the turbid milky fluid of the diseased silkworms. These are the following:

A. Streptococcus.

Morphology. Cocci perfectly round, 0,002 mm. in average diameter. (fig. 3, Pl. II.)

Agar plate. Colonies round or not perfectly round at the beginning, but later their peripheries become irregular or uneven. Color light orange, later deep orange yellow, surface more or less granulated.

Agar slant. Colony forms a light orange yellow elongated band, whose periphery is much lighter in color, and reveals a more or less serrated appearance. Medium not liquified. (fig. 1, Pl. III.)

Agar stab. Colony on the surface of medium roundish with wavy outlines. The surface of colony rough with smooth wavy periphery. That along the stab, long tubular, and becomes broader towards the surface of medium. Color light orange yellow. Medium not liquefied. (fig. 1, Pl. IV.)

Gelatine plate. Colonies roundish, light orange, later not perfectly round. The centre and periphery of the colony are deeper in color. Medium liquefied.

Gelatine stab. At the room temperature, colonies on the surface roundish, those along the stab take the shape of a long needle. Colonies light yellow. Medium liquefied. (fig. 1, Pl. V.)

Bouillon. At the room temperature, colonies are well formed and sink down to the bottom of the tube as a slight yellow precipitate.

Potato. Colony nearly round with uneven outlines. Its diameter about 10 mm. at the end of 8 days. The surface and periphery of colony show a granulated appearance. (fig. 7, Pl. II.)

Milk. Slightly coagulated near the surface of the medium, while the rest becomes more transparent. (fig. 1, Pl. VI.)

With slight acid reaction.

No indol reaction.

Stains by Gram's method, and also readily with fuchsin, gentian violet &c.

B. Streptococcus.

Morphology. Cocci perfectly round, 0,001-0,002 mm. in average diameter. (fig. 4, Pl. II.)

Agar plate. Colony roundish, light yellow with a central yellow dot. Later, it becomes lighter in color, while the periphery is irregularly serrated and retain a light yellowish color.

Agar slant. Colony light orange yellow with whitish serrated periphery, not liquefied agar. (fig. 2, Pl. III.)

Agar stab. Colony on the surface of the medium roundish with wavy outlines. Its surface more or less rough. Colonies along the stab long cylindrical and become broader near the surface. Color light greyish yellow. Not liquefied the medium. (fig. 2, Pl. IV.)

Gelatine plate. Colonies roundish, light yellow, later the periphery is undulated. Liquefied the medium.

Gelatine stab. The colony on the surface roundish, light yellow, later gelatine begins to liquefy at the surface, and assumes a funnel shape (fig. 2, Pl. V.)

Bouillon. At the room temperature, the bouillon becomes turbid, but as the yellowish precipitate sink down, it again becomes clear. On the surface of the bouillon is formed a thin film.

Potato. Colony irregular in shape. Light orange with lighter periphery. The surface with granulated appearance. (fig. 8, Pl. II.)

Milk. Slightly coagulated near the surface, and there are formed some transparent portions of the shape of cracks in the medium. (fig. 2, Pl. VI.)

With acid reaction.

No indol reaction.

Stains by GRAM's method and also readily with fuchsin, gentian violet &c.

C. Streptococcus.

Morphology. Cocci perfectly round 0.0012-0.003 mm. in average diameter. (fig. 5, Pl. II.)

Agar plate. Colony roundish, lively greenish yellow, smooth.

Agar slant. Colony dull greenish yellow. Its periphery smooth and even (fig. 3, Pl. III.)

Agar stab. Colony on the surface of the medium roundish and smooth (fig. 3, Pl. IV). Dull greenish yellow. Colonies along the stab grow in the shape of a nail.

Gelatine plate. Colony roundish, light greenish yellow. Its central portion slightly raised up, and the periphery smooth and even.

Gelatine stab. Colonies grow slower than the a and b *Streptococci*. Greenish yellow in color. Those on the surface of the medium roundish and smooth. Those along the stab, needle shaped. (fig. 3, Pl. V.)

Bouillon. At the room temperature, the medium remains clear for three days, later there may be deposited a few light greenish yellow precipitate at the bottom of the test tube. The precipitate is sticky.

Potato. Colony nearly roundish at the end of a week; but its outlines are more or less undulated. Color dull greenish yellow. Surface smooth and glossy; but more or less uneven. (fig. 9, Pl. II.)

Milk. Not coagulated. With a slight acid reaction. (fig. 3, Pl. VI.)

No indol reaction.

Stains by GRAM'S method, and also with fuchsin, gentian violet &c.

D. Streptococcus.

Morphology. Cocci nearly roundish, and some much larger in size than the above mentioned three cocci. Diameter varies from 0.001 to 0.004 mm. (fig. 6, Pl. II.)

Agar plate. Colony nearly roundish with wavy outlines. Slightly elevated at its centre, and gradually thinning out towards the periphery. Color milky white.

Agar slant. Colony milky white with more or less undulated peri-

phery. (fig. 4, Pl. III.)

Agar stab. Colonies form a milky white pellicle of a roundish shape on the surface of the medium at the end of three days. The surface of the pellicle is marked all over with a mesh work, and the periphery more or less undulated. (fig. 4, Pl. IV.)

Gelatine plate. Colonies roundish, milky white. Their central portion is slightly raised up, while the peripheries are gradually flattened.

Gelatine stab. The colonies on the surface of the medium roundish. Whitish. Those along the stab are long needle shaped. The head of the needle looks to be fringed with veils. The surface of the medium liquefies later in the shape of bowl. (fig. 4, Pl. V.)

Bouillon. At the room temperature, it becomes little turbid at the end of three days. A sticky white precipitate sinks down later to the bottom of the test tube.

Potato. Colony roundish, milky white with a light greyish shade. The surface even and somewhat glossy. (fig. 10, Pl. II.)

Milk. Coagulates, and a larger portion of the medium becomes transparent. With a strong acid reaction. (fig. 4, Pl. VI.)

No indol reaction.

The result of the infection experiments with the above mentioned four *Streptococci* upon the silkworms are as follows:—

EXPERIMENT I.

On the 7th June 1905, I took 150 worms of Akabiki race just after the fourth moult, and separated them into three lots. One of which (150 worms) was kept as a control. To one of the remaining two lots, were given the chopped leaves sprinkled over with distilled water containing *Streptococcus* (a), and to the other, chopped leaves with *Streptococcus* (b). To the control were given the chopped leaves sprinkled over simply with pure distilled water.

The results obtained were as follows:—

Date of Experiment.	Control.	Worms with Strept. (a).	Worms with Strept. (b).
7 June, 1905.	healthy.	healthy.	healthy.
8 " "	"	"	"
9 " "	"	"	"
10 " "	"	"	"
11 " "	"	"	"
12 " "	"	"	"
13 " "	"	"	"
14 " "	1 flaccid.	1 diseased.	"
15 " "	healthy.	healthy.	"
16 " "	"	"	"
17 " "	"	"	"
18 " "	"	"	"
19 " "	"	8 flaccid.	3 flaccid.
20 " "	4 flaccid.	6 "	6 "
21 " "	1 "	5 "	6 "
22 " "	4 "	1 "	4 "
	9 "	21 "	19 "
23 " "	all healthy worms in those lots began to mount.		

Of the 49 diseased worms, only one showed the symptom of Jaundice, while the remaining 48, that of flacherie; but all of the latter contained also the polyhedral bodies characteristic of Jaundice.

EXPERIMENT II.

a. On the 16th June, 1905, 150 worms of Awobiki race just after the 4th moult were taken and separated into three lots. The 1st lot of the worms was subcutaneously injected with a milky fluid taken out of the diseased worms, diluted with distilled water. The 2nd lot was treated in the same way with distilled water containing *Streptococcus* (a), while the 3rd lot with that containing *Streptococcus* (b). The results obtained were as follows:—

Date of Experiment.	Number of diseased worms.		
	1st lot.	2nd lot.	3rd lot.
16 June, 1905.	1	—	—
18 " "	1	5	3
19 " "	6	3	12
20 " "	7	19	16
21 " "	2	9	3
22 " "	22	—	1
23 " "	—	1	3
Number of dead worms during pupation.	8	3	2
	47	40	40
23 June, 1905.	all healthy worms in three lots began to mount.		

Of the 47 diseased worms of the 1st lot, 32 showed the symptom of Jaundice, while the remaining 15, that of flacherie, and the latter contained also the polyhedral bodies characteristic of Jaundice. Hence all the 47 diseased worms were affected by Jaundice.

Of the 40 diseased worms of the 2nd lot, 3 showed the symptom of Jaundice, the remaining 37 that of flacherie. Of these 37 flaccid worms, 9 contained the polyhedral bodies. So that, 12 of the 40 diseased worms were affected by Jaundice.

Of the 40 diseased worms of the 3rd lot, 4 showed the symptom of Jaundice, the remaining 36, that of flacherie, and of the 36 flaccid worms 12 contained the polyhedral bodies. So that 24 of the 40 diseased worms were affected by Jaundice.

The number of worms affected by Jaundice in each of the 3 lots of worms taken for the experiment, was as follows:—

	1st lot (50 worms).	2nd lot (50 worms).	3rd lot (50 worms).
Number of worms infested with Jaundice, that's those containing the polyhedral bodies.	47	12	24

This experiment teaches us the infective power in the 1st lot was far stronger than in the 2nd and 3rd lots.

b. On the 15th May 1907, 30 healthy worms of Awobiki race (1st day of the 4th age) were taken and subcutaneously injected with the turbid milky fluid of the diseased worms (Jaundice). The results of this experiment were:—

Date of Experiment.	Remarks.
15 May, 1907.	healthy.
16 " "	"
17 " "	"
18 " "	"
19 " "	"
20 " "	"
21 " "	"
22 " "	"
23 " "	{ all the worms affected by Jaundice.

c. On the 17th May, 1907, 30 healthy worms of Awobiki race (3rd day of the 4th age) were taken, and subcutaneously injected with the turbid milky fluid of the diseased worms (Jaundice). The results were:

Date of Experiment.	Remarks.
17 May, 1907.	healthy.
18 " "	"
19 " "	"
20 " "	"
21 " "	"
22 " "	"
23 " "	{ all the worms affected by Jaundice.

On the 8th July 1905, 300 healthy worms of Chiyodzuru race just after the 2nd moult were separated into 3 lots. The 1st lot was kept as a control, and to the 2nd were given chopped leaves bearing *Streptococcus* (a) and to the 3rd, those bearing *Streptococcus* (b). The results were as follows:—

Date of Experiment.	Number of diseased worms.		
	1st lot.	2nd lot.	3rd lot.
8 July, 1905.	—	—	—
9 " "	—	—	—
10 " "	—	1	—
11 " "	—	—	—
12 " "	—	1	1
13 " "	—	10	11
14 " "	—	9	5
15 " "	—	—	—
16 " "	—	—	—
17 " "	1	6	25
18 " "	2	—	5
19 " "	—	2	—
20 " "	—	—	—
21 " "	2	—	—
22 " "	3	—	—
	8	29	47
22 " "	all the worms began to mount.		
Number of dead worms during pupation	12	1	1
	20	30	48

Of 20 diseased worms of the 1st lot, 2 worms showed the symptom of Jaundice, while of the remaining 18 flaccid worms 5 contained the polyhedral bodies, so that in this lot, 7 of the 50 worms were affected by Jaundice.

Of the 30 diseased worms of the 2nd lot, 10 worms showed the symptoms of Jaundice, while of the remaining 20 flaccid worms 5 contained the polyhedral bodies, so that in this lot 15 of the 50 worms were affected by Jaundice.

Of the 48 diseased worms of the 3rd lot, 26 showed the symptom of Jaundice, while of the remaining 22 flaccid worms 16 contained the polyhedral bodies, so that in this lot, 42 of the 50 worms were affected by Jaundice.

EXPERIMENT IV.

On the 13th July 1905, 150 healthy worms of Chiyodzuru race (2nd breed of bivoltini) just after the 4th moult, were taken and separated into 3 lots (each containing 50 individuals). The 1st lot was kept as a control. To the 2nd, were given chopped leaves covered with distilled water containing *Streptococcus* (a). To the 3rd, were given also chopped leaves covered with distilled water containing *Streptococcus* (b). The results were as follows:—

Date of Experiment.	Number of Diseased worms.		
	1st lot.	2nd lot.	3rd lot.
13 July, 1905.	—	—	—
14 " "	—	—	—
15 " "	—	5	9
16 " "	—	6	8
17 " "	3	10	—
18 " "	—	—	12
19 " "	—	—	2
20 " "	—	1	2
21 " "	—	—	2
Number of dis- eased worms } during pupation.	4	1	3
	7	23	38

Of the 7 diseased worms of the 1st lot 6, of the 23 diseased ones of the 2nd lot 10, and of the 38 diseased worms of the 3rd lot 13 were affected by Jaundice.

EXPERIMENT V. (Repetition of Exp. IV.)

On the 10th May 1907, 120 healthy worms of Awobiki race (5th day of 3rd age) were selected and separated into 4 lots.

To the 1st lot (kept as control) were given normal chopped leaves, while to the 2nd, 3rd and 4th, chopped leaves bearing respectively, *a*, *b*, and *c Streptococci*. The results were:—

Date of Experiment.	Number of diseased worms.			
	1st lot.	2nd lot.	3rd lot.	4th lot.
10 May, 1907.	—	—	—	—
11. " "	—	—	—	—
12. " "	—	—	—	—
13. " "	—	—	—	—
14. " "	—	—	—	—
15. " "	—	—	—	—
16. " "	—	—	—	—
17. " "	—	—	—	—
18. " "	—	—	—	—
19. " "	—	—	—	—
20. " "	1	—	—	1
21. " "	—	—	—	—
22. " "	1	—	—	—
23. " "	—	—	—	—
24. " "	—	—	—	—
25. " "	—	—	—	—
26. " "	—	—	—	—
27. " "	—	—	—	—
28. " "	—	—	—	1
29. " "	Mounted.	Mounted.	Mounted.	Mounted.
5-6 June, "	6 larvae died in the cocoon.	10 larvae 4 pupa died in the cocoon.	7 larvae died in the cocoon.	9 larvae died in the cocoon.

In the first lot, 2 diseased worms appeared during cultivation, one of which suffered from Jaundice, and of the 6 larvae that died in the cocoon, 4 contained the polyhedral bodies, so that there were 5 worms affected by Jaundice.

In the 2nd lot, of the 10 larvae that died within the cocoon, 9 contained the polyhedral bodies.

In the 3rd lot, of the 7 worms that died within the cocoon, 6 contained the polyhedral bodies.

In the 4th lot, of the 9 larvae that died within the cocoon, 4 contained the polyhedral bodies.

Thus the number of the worms affected by Jaundice in the above mentioned 4 lots is shown for convenience in the following table:—

Nos. of lots.	Nos. of worms taken for experiments.	Nos. of worms affected by Jaundice.
1	30	5
2	30	9
3	30	6
4	30	4

EXPERIMENT VI.

I. On the 18th May 1907, 60 healthy worms of Awobiki race (3rd day of the 4th age) were taken, and separated into 3 lots (each of 30 worms). Each of them was subcutaneously injected with the *Streptococci* (a) (b) (c), which have been procured from the turbid milky fluid of the diseased worms (affected with Jaundice).

1st lot. 20 worms were subcutaneously injected with the *Streptococcus* (a). The results were:—

Date of Experiment.	Remarks.
18 May, 1907.	healthy.
19 " "	"
20 " "	"
21 " "	4th moult.
22 " "	healthy.
23 " "	"
24 " "	"
25 " "	2 flaccid in which 1 contained polyhedral bodies.
26 " "	healthy.
27 " "	"
28 " "	4 flaccid in which 3 contained polyhed. bodies.
29 " "	healthy.

Date of Experiment.	Remarks.
30 May, 1907.	4 flaccid in which 2 contained polyhed. bodies.
31 " "	healthy.
1 June, "	4 flaccid in which 3 contained polyhed. bodies.
2 " "	mounted.
3 " "	3 flaccid in which 2 contained polyhed. bodies.
<hr/>	
17 flaccid. 11 Jaundice.	

In this experiment, there were 17 flaccid worms, but none silkworms showed the symptom of Jaundice; but on examining the turbid blood of 11 Jaundice all contained the characteristic polyhedral bodies.

2nd lot. 20 worms were subcutaneously injected with the *Streptococcus* (b). The results were:

Date of Experiment.	Remarks.
18 May, 1907.	healthy.
19 " "	"
20 " "	"
21 " "	"
22 " "	"
23 " "	"
24 " "	"
25 " "	"
26 " "	"
27 " "	"
28 " "	4 flaccid in which 3 contained polyhed. bodies.
29 " "	healthy.
30 " "	2 flaccid.
31 " "	healthy.
1 June, "	"
2 " "	8 mounted.
3 " "	2 flaccid.
4 " "	3 flaccid in which 3 contained polyhed. bodies.
<hr/>	
11 flaccid. 6 Jaundice.	

In this experiment, I found 11 flaccid worms, but none showed the symptoms of Jaundice; but on examining the turbid blood of the flaccid silkworms, I found in 6 individuals the characteristic polyhedral bodies.

3rd lot. 20 worms were subcutaneously injected with *Streptococcus* (c). The results were:—

Date of Experiment.		Remarks.
18	May, 1907.	Nos. of diseased worms.
19	" "	healthy.
20	" "	"
21	" "	"
22	" "	"
23	" "	"
24	" "	"
25	" "	"
26	" "	"
27	" "	"
28	" "	"
29	" "	"
30	" "	"
31	" "	"
1	June, "	19 worms mounted.
2	" "	no diseased worms.
3	" "	"
4	" "	"

By the above experiments, it is concluded that the silkworms inoculated with the *Streptococci* (a) and (b) show the symptoms of Jaundice, while those inoculated with (c) do not.

EXPERIMENT VII.

On the 2nd June 1908, 100 healthy worms of Awobiki race (1st day of 5th age) were taken and separated into 5 lots (each 20 individuals). The first four lots were subcutaneously inoculated with the *Streptococci* (a, b, c, d), and the 5th lot was normally fed for control. The results of this experiment were as follows:

a. On the 2nd June 1908, the 1st lot of Awobiki race subcutaneously injected with the *Streptococcus* (a). The results were:—

Date of Experiment.		Remarks.
2	June, 1908.	healthy.
3	" "	"
4	" "	"
5	" "	"
6	" "	"
7	" "	"
8	" "	"
9	" "	"
10	" "	all worms mounted.

After mounting, 2 worms died in the cocoonery and 6 worms within the cocoon; so that there were 8 dead worms in this lot in all. These 8 worms contained *Streptococcus* (a) and two of them also the polyhedral bodies.

b. On the 2nd June 1908, the 2nd lot of Awobiki race subcutaneously injected with *Streptococcus* (b). The results were:—

Date of Experiment.	Remarks.
2 June, 1908.	healthy.
3 " "	"
4 " "	"
5 " "	"
6 " "	"
7 " "	"
8 " "	1 flaccid.
9 " "	healthy.
10 " "	mounted.

After mounting, 3 worms died in the cocoonery and 1 within the cocoon. Of the 4 dead worms, 2 contained the polyhedral bodies besides *Streptococcus* (b).

c. On the 2nd June 1908, the 3rd lot of Awobiki race subcutaneously injected with *Streptococcus* (c). The results were:—

Date of Experiment.	Remarks.
2 June, 1908.	healthy.
3 " "	"
4 " "	"
5 " "	"
6 " "	"
7 " "	1 flaccid.
8 " "	healthy.
9 " "	"
10 " "	19 mounted.

After mounting, 3 worms died within the cocoon, these contained only *Streptococcus* (c).

d. On the 2nd June 1908, the 4th lot of Awobiki race subcutaneously injected with *Streptococcus* (d). The results were:—

Date of Experiment.	Remarks.
2. VI, 1908.	healthy.
3 " "	"
4 " "	"
5 " "	"
6 " "	"
7 " "	"
8 " "	"
9 " "	"
10 " "	20 worms mounted.

After mounting, one worm died in the cocoonery, and 4 worms, within the cocoon.

Of the 5 dead worms, 3 contained the polyhedral bodies besides *Streptococcus* (*d*).

e. On the same date, the 5th lot of Awobiki race was taken as control. The results were:—

Date of Experiment.	Remarks.
2 June, 1908.	healthy.
3 " "	"
4 " "	"
5 " "	"
6 " "	"
7 " "	"
8 " "	"
9 " "	"
10 " "	20 worms mounted.

After mounting, 1 worm died in the cocoonery and 2, within the cocoon. All these three dead worms did not contain the polyhedral bodies.

EXPERIMENT VIII.

On the 17th June 1908, 100 healthy worms of Awobiki race (2nd day of 5th age) were taken, and separated into 5 lots (each embraced 20 individuals). To the first 4 lots were subcutaneously injected with *Streptococci* (*a*) (*b*) (*c*) and (*d*) respectively. The remaining lot was treated as control.

The experiments were carried on as follows:—

a. On the same date, the 1st lot of Awobiki race subcutaneously injected with *Streptococcus* (a). The results were:—

Date of Experiment.	Remarks.
17 June, 1908.	healthy.
18 " "	"
19 " "	"
20 " "	"
21 " "	"
22 " "	7 flaccid.
23 " "	13 mounted.

After mounting, 3 worms died in the cocoonery, and 6, within the cocoon. Of the 9 dead worms, 4 contained the polyhedral bodies besides *Streptococcus* (a).

b. On the same date, the 2nd lot of the same race subcutaneously injected with *Streptococcus* (b). The results were:—

Date of Experiment.	Remarks.
17 June, 1908.	healthy.
18 " "	"
19 " "	"
20 " "	1 flaccid.
21 " "	healthy.
22 " "	"
23 " "	19 mounted.

After mounting, 4 worms died in the cocoonery, and 7, within the cocoon. Of the 12 dead worms, 5 contained the polyhedral bodies besides *Streptococcus* (b).

c. On the same date, the 3rd lot of the same race subcutaneously injected with *Streptococcus* (c). The results were:—

Date of Experiment.	Remarks.
17 June, 1908.	healthy.
18 " "	"
19 " "	"
20 " "	2 flaccid.
21 " "	1 "
22 " "	1 "
23 " "	16 mounted.

After mounting, 3 worms died in the cocoonery and 6, within the cocoon. Of the 13 dead worms, 4 contained the polyhedral bodies besides *Streptococcus* (c).

d. On the same date, the 4th lot of the same race subcutaneously injected with *Streptococcus* (d). The results were:—

Date of Experiment.	Remarks.
17 June, 1908.	healthy.
18 " "	"
19 " "	"
20 " "	1 flaccid.
21 " "	healthy.
22 " "	"
23 " "	19 mounted.

After mounting, 5 worms died in the cocoonery, and 5, within the cocoon. Of the 11 dead worms, none contained the polyhedral bodies.

e. The control lot gave the following results:—

Date of Experiment.	Remarks.
17 June, 1908.	healthy.
18 " "	"
19 " "	"
20 " "	"
21 " "	"
22 " "	"
23 " "	20 mounted,

After mounting, 1 worm died in the cocoonery, and 2, within the cocoon. The 3 dead worms contained no polyhedral bodies.

EXPERIMENT IX.

On the 17th June 1908, again 100 healthy worms of Awobiki race (5th day 5th age) were taken and separated into 5 lots (each of 20 individuals). The first four lots were subcutaneously injected with *Streptococci* (a), (b), (c), (d) respectively. A remaining lot was treated as control.

The experiments were carried on as follows:—

b. On the 17th June 1908, the first lot of the Awobiki race subcutaneously injected with *Streptococcus* (a). The results were:

Date of Experiment.	Remarks.
17 June, 1908.	healthy.
18 " "	"
19 " "	"
20 " "	20 mounted

After mounting, 4 worms died within the cocoon, 2 of them contained the polyhedral bodies in addition to the *Streptococcus* inoculated.

b. On the same date, the 2nd lot of the same race subcutaneously injected with *Streptococcus* (b). The results were:—

Date of Experiment.	Remarks.
17 June, 1908.	healthy.
18 " "	"
19 " "	1 flaccid.
20 " "	19 mounted.

After mounting, 2 worms died within the cocoon. All the dead worms contained the polyhedral bodies.

c. On the same date, the 3rd lot of the same race subcutaneously injected with *Streptococcus* (c). The results were:—

Date of Experiment.	Remarks.
17 June, 1908.	healthy.
18 " "	"
19 " "	"
20 " "	1 flaccid, 19 mounted.

After mounting, one worm died in the cocoonery and 4, within the cocoon. Two of the 6 dead worms contained the polyhedral bodies in addition to *Streptococcus* inoculated.

d. On the same date, the 4th lot of the same race subcutaneously injected with *Streptococcus* (d). The results were:—

Date of Experiment.	Remarks.
17 June, 1908.	healthy.
18 " "	"
19 " "	1 flaccid.
20 " "	19 mounted.

After mounting, one worm died in the cocoonery, and 6, within the cocoon. Of the 8 dead worms 4 contained the polyhedral bodies in addition to *Streptococcus* inoculated.

e. The 5th (control) lot gave the following:—

Date of Experiment.	Remarks.
17 June, 1908.	healthy.
18 " "	"
19 " "	"
20 " "	20 mounted.

After mounting, 2 worms died in the cocoonery, and 2, within the cocoon. All the 4 dead worms contained no polyhedral bodies.

EXPERIMENT X.

On the 19th June 1908, 100 healthy worms of Awobiki race (1st day 5th age), were taken and separated into 5 lots (each of 20 individuals). The first four lots were subcutaneously injected with *Streptococci* (a), (b), (c), (d) respectively. The remaining lot was treated as control. The experiments were carried on as follows:

a. On the 19th June 1908, the 1st lot of Awobiki race subcutaneously injected with *Streptococcus* (a). The results were:

Date of Experiment.	Remarks.
19 June, 1908.	healthy.
20 " "	"
21 " "	5 flaccid.
22 " "	5 flaccid.
23 " "	healthy.
24 " "	"
25 " "	1 flaccid.
26 " "	9 mounted.

After mounting, 4 worms died in the cocoonery. Of 15 dead worms, 4 contained the polyhedral bodies.

b. On the same date, the 2nd lot of the same race subcutaneously injected with *Streptococcus* (b). The results were:

Date of Experiment.	Remarks.
19 June, 1908.	healthy.
20 " "	"
21 " "	"
22 " "	2 flaccid.
23 " "	4 "

Date of Experiment.	Remarks.
24 June, 1908.	2 flaccid.
25 " "	1 "
26 " "	11 mounted.

After mounting, 3 worms died in the cocoonery, and 3 within the cocoon. None of the 15 dead worms contained the polyhedral bodies.

c. On the same date, the 3rd lot of the same race subcutaneously injected with *Streptococcus* (c). The results were:—

Date of Experiment.	Remarks.
19 June, 1908.	healthy.
20 " "	"
21 " "	3 flaccid.
22 " "	17 "
23 " "	—
24 " "	—
25 " "	—
26 " "	—

None of the dead worms contained the polyhedral bodies.

d. On the same date, the 4th lot of the same race subcutaneously injected with *Streptococcus* (d). The results were:

Date of Experiment.	Remarks.
19 June, 1908.	healthy.
20 " "	"
21 " "	9 flaccid.
22 " "	11 "

Of the 20 dead worms, 4 contained the polyhedral bodies in addition to *Streptococcus* (d).

e. The 5th lot kept for control gave the following results:

Date of Experiment.	Remarks.
19 June, 1908.	healthy.
20 " "	"
21 " "	"
22 " "	"
23 " "	1 flaccid.
24 " "	2 "
25 " "	17 mounted.

After mounting, a worm died within the cocoon, none of 4 dead worms contained the polyhedral bodies.

The Relation of Formalin and Certain Food Plants to Jaundice.

a. Formalin. In 1907, Messrs. D. HAYASHI³² and T. NAKAJIMA first informed us, that the Jaundice may be caused by giving formalin to the silkworms, but they did not describe the details of the process. To determine whether their experiments are correct or not, I repeated very carefully the same experiments by giving formalin of various percentage to the silkworms.

1. On the 12th June 1907, 6 lots (each of 20 healthy worms) of the silkworms of Awobiki race (1st day of the 5th age) were selected, and the food leaves sprinkled over with 2%, 4%, 6%, 8%, 10% and 15% formalin were given to them. The results were as follows:—

Date of Experiment.	2% Formalin.	4% Formalin.	6% Formalin.	8% Formalin.	10% Formalin.	15% Formalin.
12 June, 1907.	healthy.	healthy.	healthy.	healthy.	healthy.	healthy.
13 " "	"	"	"	"	"	"
14 " "	"	"	"	"	5 diseased.	"
15 " "	"	"	"	"	5 " "	"
16 " "	"	"	"	"	healthy.	2 diseased.
17 " "	4 diseased.	2 diseased.	"	"	"	3 " "
18 " "	1 " "	5 " "	6 diseased.	5 diseased.	"	healthy.
19 " "	healthy.	5 " "	7 " "	10 " "	9 diseased.	15 diseased.
20 " "	"	healthy.	7 " "	5 " "	1 lost.	—
21 " "	"	"	—	—	—	—

5 diseased. 12 diseased. all diseased. all diseased. 19 diseased. all diseased.

From this, we see that the formalin of 4-15% is an effectual agent in producing Jaundice.

2. On the 31st, May, 1907, 4 different sorts of diluted formalin (1-4%) were prepared, and each subcutaneously injected to each (30

32. Report of the Sericultural Association of Japan (Japanese). No. 180. p. 1—3. 1907.

silkworms) of the 4 lots of the healthy silkworms of Awobiki race (3rd day 4th age). The results were:—

Date of Experiment.	1% Formalin.	2% Formalin.	3% Formalin.	4% Formalin.
1 June, 1907.	healthy.	healthy.	healthy.	healthy.
2 " "	"	"	"	"
3 " "	"	"	"	"
4 " "	"	"	"	"
5 " "	"	"	"	"
6 " "	"	"	"	1 flaccid.
7 " "	"	"	"	healthy.
8 " "	"	"	"	1 flaccid.
9 " "	1 flaccid.	"	"	healthy.
10 " "	healthy.	"	"	"
11 " "	3 flaccid.	1 flaccid.	"	"
12 " "	healthy.	healthy.	"	2 flaccid.
13 " "	"	"	1 flaccid.	healthy.
14 " "	26 mounted.	29 mounted.	29 mounted.	26 mounted.

3. On the 16th June 1907, 80 healthy silkworms of Awobiki race (2nd day 5th age) were taken, and separated into 5 lots, to each of which formalin was subcutaneously injected in the strengths of 0.1%, 0.2%, 0.5% and 0.7% respectively. The results were:—

Date of Experiment.	0.1% Formalin.	0.2% Formalin.	0.5% Formalin.	0.7% Formalin.
6 June, 1907.	healthy.	healthy.	healthy.	healthy.
7 " "	"	"	"	"
8 " "	"	"	"	"
9 " "	"	"	"	"
10 " "	"	"	"	"
11 " "	"	"	"	"
12 " "	"	"	"	"
13 " "	"	"	"	"

A comparison of these results will show that formalin less than 1% has no power to produce the Jaundice, while in strength greater than 1% it will produce the disease more or less, and when it is stronger than 4%, all the worms are affected and will die.

4. At 11 a.m. 10th May 1908, three lots (each of 30 individuals) of the healthy worms of Koishimaru race (1day 3rd age) were selected. To the first two lots were given chopped leaves sprinkled over with

formalin (4 %), and the remaining one was kept for control and fed with ordinary chopped leaves. The results obtained were:—

Date of Experiment.	1st lot. (leaves with 4% formalin given.)	2nd lot. (leaves with 4% formalin given.)	3rd lot. (control).	Remarks.
10 May, 1908.	—	—	—	
11 „ „	—	—	—	
12 „ „	—	—	1 flaccid.	
13 „ „	—	—	—	
14 „ „	—	—	—	
15 „ „	—	—	—	
16 „ „	5 diseased.	—	—	
17 „ „	3 „	3 diseased.	—	ended 4th moult.
18 „ „	—	1 „	—	
19 „ „	1 diseased.	1 „	—	
20 „ „	—	—	—	
21 „ „	—	—	—	
22 „ „	—	—	—	
23 „ „	—	—	—	
24 „ „	—	—	—	
25 „ „	—	—	—	
	9 diseased.	5 diseased.	1 diseased.	

5. At 1 p.m. 15, May 1908, three lots (each at 20 individuals) of the healthy worms of Matamukashi race (1st day 3rd age) were selected. To the first lot were given chopped leaves sprinkled over with 8% formalin, and to the third ordinary chopped leaves. The results obtained were:—

Date of Experiment.	1st lot. (8% formalin).	2nd lot. (10% formalin).	3rd lot. (control).
15 May, 1908.	10 died.	17 died.	—
16 „ „	6 „	1 „	—
17 „ „	1 „	—	—
18 „ „	—	—	—
19 „ „	—	2 flaccid.	—
20 „ „	—	—	1 diseased.
21 „ „	4 diseased.	—	—
22 „ „	—	—	—
5 June, „	—	—	19 mounted.
8 „ „	—	—	2 worms died within th cocoon.

In this experiment, 4 diseased worms in the 1st lot, 2 flaccid in the

2nd lot, and a dead worm within the cocoon contained the polyhedral bodies.

6. At 4 P.M. 15 May 1908, five lots (each of 20 individuals) of the healthy worms of Awobiki race (4th day 5th age) were selected. To the first lot were given chopped leaves sprinkled over with 15% formalin; to the second, those with 20%; to the third, those with 36%, and to the fifth (control), ordinary chopped leaves without formalin. The results were:—

Date of Experiment.	1st lot.	2nd lot.	3rd lot.	4th lot.	5th lot.
15 June, 1908.	—	9 flaccid.	2 died.	10 flaccid.	—
16 „ „	—	—	5 flaccid.	7 „	—
17 „ „	—	—	10 „	1 „	—
18 „ „	4 diseased.	—	—	—	—
19 „ „	1 flaccid.	—	—	—	—
20 „ „	15 mounted. (13 died within the cocoon).	11 mounted. (8 died within the cocoon).	3 mounted. (2 died within the cocoon).	2 mounted. (2 died within the cocoon).	19 mounted. (5 died within the cocoon).

In the 1st lot, 17; in the 2nd, 8; in the third, 5; in the 4th, 6 dead worms contained the polyhedral bodies, and in the 5th lot there were 4 dead worms containing them. This experiment teaches us that the formalin produces more or less Jaundice; and the Jaundice in the 5th lot appears to have been produced in worms infested by maggots.

b. On the 22nd May 1907, 20 healthy silkworms of Akabiki race (1st day 4th age) were taken and fed with the leaves of *Cudrania triloba*, which are employed largely in China as the food-plant of silkworms. The results were:—

Date of Experiment.	Remarks.
22 May, 1907.	healthy.
23 „ „	—
24 „ „	—
25 „ „	—
26 „ „	—
27 „ „	—
28 „ „	ended 4th moult.
29 „ „	healthy.
30 „ „	1 diseased.
31 „ „	1 flaccid.

Date of Experiment.	Remarks.
1 June, „	healthy.
2 „ „	„
3 „ „	„
4 „ „	„
5 „ „	28 mounted.

This experiment shows us that the leaves of *Cudrania triloba* have relatively little power to cause Jaundice; but when the newly hatched silkworms are fed with the same leaves large numbers of them may be affected by Jaundice.

c. On the same date, 20 healthy silkworms of Akabiki race (1st day 4th age) were taken and fed with the leaves of *Broussonetia papyrifera* Vent. The results were:—

Date of Experiment.	Remarks.
22 May, 1907.	healthy.
23 „ „	„
24 „ „	„
25 „ „	„
26 „ „	„
27 „ „	„
28 „ „	ended 4th moult.
29 „ „	2 flaccid.
30 „ „	14 „
31 „ „	healthy.
1 June, „	„
2 „ „	„
3 „ „	„
4 „ „	2 Jaundice.
5 „ „	3 flaccid.

In this experiment, 2 worms died from Jaundice and the remaining 18 became flaccid, but all the latter contained the polyhedral bodies characteristic of Jaundice, so that all the 20 worms used for the experiment regarded as affected by Jaundice.

d. On the 22nd May 1907, 20 healthy worms of Akabiki race (1st day 4th age) were selected and fed with the leaves of *Taraxacum officinale* Wigg. var. *corniculatum*, and the same experiment was repeated on the 7th June 1907. The results were:—

Date of Experiment.	Remarks.
22 May, 1907.	healthy.
23 " "	"
24 " "	"
25 " "	"
26 " "	"
27 " "	7 flaccid.
28 " "	healthy.
29 " "	10 flaccid.
30 " "	healthy.
31 " "	"
1 June, "	3 diseased.

In this experiment, 3 diseased worms showed the symptom of Jaundice, while 13 of 17 flaccid worms contained the polyhedral bodies, so that 16 worms were affected by Jaundice.

The experiment on the 7th June 1907, was carried on with 20 worms of Awobiki race (2nd day 5th age). The results were:—

Date of Experiment.	Remarks.
7 June, 1907.	healthy.
8 " "	"
9 " "	"
10 " "	"
11 " "	"
12 " "	"
13 " "	5 flaccid.
14 " "	10 "

The worms which died by flaccidity, contained in every case the polyhedral bodies characteristic of Jaundice.

e. On the 4th May 1908, 2,000 newly hatched silkworms of Akabiki race were taken and they are separated into two lots (each of 1,000 worms). To the first lot were given mulberry leaves, and to the second the leaves of *Cudrania triloba*. The worms of the two lots matured, began to spin cocoons on the 7th June. The numbers of the diseased worms, which appeared during the 5 ages of the two lots and of the healthy worms which spun the cocoons were as follows:—

Ages.	Number of diseased worms of 1st lot. (1000 worms fed with mulberry leaves).		Number of diseased worms of 2nd lot. (1000 worms fed with <i>C. triloba</i>).	
	Worms infested with Jaundice.	Worms infested with other diseases.	Worms infested with Jaundice.	Worms infested with other diseases.
1st age.	0	0	0	0
2nd „	21	14	54	10
3rd „	30	12	40	15
4th „	20	4	45	5
5th „	44	25	400	20
After mounting.	20	2	151	0
Total number.	135	57	690	50

The above table shows us that there were produced 135 diseased worms (Jaundice) in the 1st lot, while in the second lot 690 diseased ones. The ratio of the diseased worms in the first and in the second lots is nearly 1 to 5, thus the worms fed with *Cudrania triloba* have a greater tendency to be subject to Jaundice than those fed with mulberry leaves.

The Interruption of Respiration in Relation to Jaundice.

The experiment of the diseased worms (Jaundice) by the interruption of respiration was done by myself about twenty years ago, and the results were described in my pamphlet entitled "Nosan" (Japanese) published in 1886. Some authors³² have criticized my opinion on the production of the diseased worms by this interruption; but my repeated experiments on the same subject in 1907 have given the same results as in the year 1886. The results of the experiments carried on 1907 are described as follows:—

a. 3 P.M. 20 June 1907, 10 worms of Awobiki race (6th day 5th age) were taken, and all the spiracles were closed up with a mixture

32. MM. A. CONTE et D. LEVRAT: Les Maladees du Ver a Soie. Laboratoire d'Études de la Soie 1906—1907. Vol. XIII.

of equal parts of vaselin and bee's wax. On the 21st June, the body of all the worms was exceedingly swollen up, and they died at 5 P.M. of the same day. None of the dead worms contained the polyhedral bodies.

b. 3 P.M. 20 June 1907, 10 worms of the same race were taken, and all the spiracles on one side of the body were closed up with the same mixture.

On the 23rd June, the body of all the worms were bent more or less on one side, and the side of which the spiracles were closed up, was swollen up. On the 24th 6 worms died. They contained more or less the polyhedral bodies within their body.

c. At the same hour and date, 10 worms were taken, and 3 spiracles on each side of the body were closed up with the same mixture. On the 20th to 22nd June, all the worms were healthy, but on the 23rd, 2 flaccid worms appeared, but showed no symptoms of Jaundice.

d. At the same hour and date, 10 worms were taken, and the same number of spiracles as in c, closed up. On the 20th to 22nd, all the worms were healthy. On the 23rd, three worms, after spinning the cocoon a single worm died. All these four dead worms contained the polyhedral bodies characteristic to Jaundice.

Camphor Odor in Relation to Jaundice.

In order to determine whether the camphor odor will become a cause for the production of the Jaundice, I took 3 lots (each of 10 individuals) of healthy silkworms (Awobiki race), and fed each of the lots separately in a bell jar (27 cm. in diameter and 9 cm. in height) containing certain amounts of camphor. All the worms died after certain lapse of time. The results were:—

Date of Experiment.	Number of lots.	Amount of camphor given.	Duration of life after the application of camphor.
7 June 1907.	1st lot.	1.1 grms.	20 hours.
	2nd lot.	1.95 „	25 „
	3rd lot.	0.5 „	2 days 4 hours.

All the dead worms contained more or less the polyhedral bodies in the blood as well as in other organs and tissues.

Production of Jaundice by the Cross of the Domestic and Wild Silkworms.

Dr. T. Toyama, who has been making studies, since 1900, on the hybridology of insects, has kindly informed me that the cross breed of the domestic and wild silkworms was always strongly affected by Jaundice. The results of his observation were as follows:—

Times reared.	Crosses.		Nos. of diseased silkworms less affected.
	♀ Awobiki race.	♂ W. S.	
1900			
1905	W. S.	Quadrivoltini race.	" "
"	"	Matamukashi race.	" "
"	"	Chinese race.	" "
1908	Himeko race. (bivoitini).	W. S.	all affected.
"	Araya race. (bivoltini).	"	" "
"	"	"	" "
"	Hihaku race	"	" "
"	Quadrivoltini race.	"	" "
1909	Himeko race.	"	90% affected.

The above table teaches us that the ratio of the production of the Jaundice during the breeding, was much greater in the cross of ♀ domestic and ♂ wild silkworms* than in that of ♀ wild and ♂ domestic silkworms.

It is quite difficult at present to answer the question why the cross breed was so strongly affected by Jaundice; but this seems, I believe, to be due to certain internal alternations resulting from the crossing somewhat similar to what occurs when the normal food plant is replaced by another. And at the same time, I should say the Jaundice produced in the cross breed is not due to the presence of parasitic micro-organisms.

W.S.=Wild Silkworm (*Theophila mandalina*).

* The wild silkworm (*Theophila mandalina* race) was first pointed out by me in 1898 as the ancestral form of the domestic silkworms. Anno. Zool. Jap. Vol II, Pars. II, 1898.

Presence of the Polyhedral Bodies in Certain Diseased Worms.

a. In Japan, the worms of the spring breed are yearly infested in large numbers by a maggot (larva of *Ugimyia* (*Crossocosmia*) *sericariae*). The worms thus infested lose their appetite, become inactive, and in many cases, some or nearly all the segments of the body swell up and the worms finally die. In such diseased worms, the blood becomes milky white and turbid. Further in the dead worms or pupae within cocoons, and particularly in those infested by the maggot, the blood contains always more or less polyhedral bodies, so that the presence of the parasitic maggot in the worms as well as in the pupae must be regarded as one of the sources for the production of Jaundice.

b. The so called Nemuradzu or Hikaruko in Japan, are the diseased worms at the periods of moultings. They are the worms, which can not undergo moultings at a due time. The skin of the body is fully expanded, and looks pale white and glossy. Their blood becomes always turbid, and is of a milky white color. Their turbidity is always due to the presence of the polyhedral bodies.

c. Many of the worms infested by flacherie contain also more or less the polyhedral bodies without relation to the turbidity of the blood.

As I have already stated before, if we assume the presence of the polyhedral bodies as the chief indication of Jaundice, all the diseased worms containing the bodies without regard to the turbidity of blood should be looked upon as subject to Jaundice.

Nature of the Polyhedral Bodies Characteristic of Jaundice.

About the nature of the polyhedral bodies in the worms affected by Jaundice (Gelbsucht, Grasserie), there are different opinions, and the question still appears to be undecided, and therefore it will not be entirely useless to discuss it.

The polyhedral bodies usually do not easily take up stains; but when they are heated on the flame of an alcohol lamp or in a steam for 30 minutes to one hour, they become very readily stainable with various colouring matters, particularly such as hæmatoxylin, picric acid, carbol fuchsin, and other aniline colors. They are tinged brown by osmic acid, and yellowish brown by potassic iodide or iodine.

As director BOLLE has stated before, the polyhedral bodies are easily crushed by pressure, viz. when we put a cover glass on a glass plate, on which a small drop of blood containing them, is placed, and press strongly the cover glass by means of a needle, all of the bodies are broken into small pieces, or become irregular in shape by the production of radial cracks or streaks. This indicates the brittleness of the polyhedral bodies. The same effect is produced when the bodies are dried up between a glass slide and cover glass, or when they are exposed to the temperature of 120° C. for 20 hours, or 140° C. for 10 hours. Even after the exposure of the bodies to the above temperatures, some of them retain still their polyhedral shape.

The polyhedral bodies have a strong resisting power against several reagents, and in most cases do not alter their shape by the addition of several substances. Thus:

1. The polyhedral bodies, which have been preserved in 90% alcohol, when examined after three years or even more do not show any change of shape.

2. Caustic potash (10%) does not act upon the polyhedral bodies, but its concentrated solution dissolves them immediately. Chloroform, ether, acetic acid and even sulphuric, nitric, hydrochloric acids &c. do not act upon them; but the polyhedral bodies become more clear and transparent than before.

3. In saliva, the polyhedral bodies do not change their shape, but if it dries up, they are crushed into some irregular pieces as in the exposure in the air.

The same bodies, which are kept in the blood or gastric juice of silkworms, remain unaltered even after three months.

On the 27th May 1900, the polyhedral bodies were placed in a hanging drop of bouillon prepared from silkworm extracts, and I have examined them at certain intervals till 19 July 1900; but I could not find any change.

On the 20th May 1907, the same bodies were transferred into glycerine-water (equal part of glycerine and water), salt solution (5%), bouillon &c. No change after ten days.

From the experiments with bouillon, glycerine water, and salt solution, it is clear that the polyhedral bodies are not a sort of spore of certain lower organisms as was supposed to be by director J. BOLLE, who proposed the name of *Microsporidium polyedricum*.

It is generally admitted that the blood of the diseased worms (Jaundice) becomes turbid and is of either whitish or yellowish color according as the race is white or yellow. Certainly, the turbidity of the blood is a clear and distinct symptom of the Jaundice, but very frequently, it does not undergo this change remaining clear as in the healthy worms. And therefore the turbidity of the blood can not be considered as the main symptom of the disease; but the presence of the polyhedral bodies without regard to the quality of the blood, in the organs or tissues of the worm must be looked upon as a distinct indication of this disease.

Now, if we examine the diseased worms, we can always detect the polyhedral bodies also in the walls of the stomach, dorsal vessel, trachea, hyperdermal layer of skin, malpighian vessel, adipose tissue, silk glands, &c. (figs. 1-6, Pl. VI.)

If we prepare the sections of the above mentioned organs or tissues hardened in Frenzel's solution and colored with acid fuchsin, picric acid or with hæmatoxylin and fuchsin, then we can easily find in many of the tissue cells well stained polyhedral bodies of various sizes.

These bodies can be distinctly seen particularly in the cells of the fatty tissue, hyperdermal layer of the skin &c. (figs. 1-6, Pl. VI.) In the cells of the fatty tissue, we can always detect the polyhedral bodies with great ease. Some of the cells may be seen to contain certain number of minute dots or granules in their nucleus, while in others, the polyhedral

bodies of various sizes. These granules, though very minute, have a glassy nature exactly similar to the polyhedral bodies, and colored exactly same as the latter by the same coloring matters. Moreover, there may be usually seen intermediate stages between these granules and the distinct polyhedral bodies. This fact will prove that these distinct polyhedral bodies are derived from these glassy dots or granules.

The number of these granules in a single cell is usually not larger than that of the larger polyhedral bodies. If these granules were formed after successive division of the larger polyhedral bodies, the number of the former in a single cell must be greater and fill up the nucleus; but this is not usually met with, and the granules lie comparatively few and scattered in the nucleus of the cell. This shows us that the granules are not produced as the result of division of the distinct larger polyhedral bodies. Thus we are inclined to believe that the glassy granules are merely products formed inside the nucleus, due to certain unknown process, and that they are not spores or some developmental stages of certain micro-organisms, as director J. BOLLE stated in 1898.

The nuclear membrane of the nucleus containing the glassy granules as well as the polyhedral bodies, looks in general to be firmer than that of the normal nucleus, and furthermore, the nucleus containing the granules or polyhedral bodies grows gradually larger as if owing to their presence in the cell. As a result of its growth, the nucleus gets free by breaking up the cell wall in which it is enclosed. The free nucleus is thrown into the blood, and circulates for some time. This is unquestionably nothing else than those which are stated by director J. BOLLE as a cyst of a sort of Sporozoa (*Microsporidium polyedricum*). It is very often the case, that as the nucleus containing the polyhedral bodies grows in excess, the nuclear membrane breaks up just before or after the rupture of the cell walls. (fig. 6, Pl. VI.) In such cases, large numbers of the polyhedral bodies are thrown into the blood so as to render the latter turbid. Sometimes the nucleus, which is filled up with the polyhedral bodies without rupture of its membrane, gets out of the cell,

and swims in the blood. It seems that what director J BOLLE³³ calls "Bläschen" is nothing else than the above mentioned nucleus. Sometimes, the blood corpuscles of the silkworms take in some of the polyhedral bodies, when they look also as a sort of "Bläschen."

As to the origin of the polyhedral bodies, there are different opinions. Messrs F. A. WACHTL and K. KORNAUTH,³⁴ who studied the "Wipfelkrankheit" of *Psilura monacha* L. have compared it with the Jaundice of the silkworm, and stated its cause as follows: "Das charakteristische für die Wipfelkrankheit der Nonnenraupe ist also das Auftreten dieser ganz eigenthümlich geformten Körnchen und Fehlen von grösseren Bakterienmassen. . . . Zuerst scheinen die Körnchen in Fettkörper aufzutreten, welchen sie bald vollständig erfüllen. Später erscheinen sie vereinzelt im Blute, und zwar in den Blutkörperchen, dieselben schliesslich ebenfalls vollständig erfüllend. Nach dem Bersten des Blutkörperchen treten die Körnchen aus und schwimmen nun frei in der Blufflüssigkeit umher. Zugleich findet Man sie auch in nahezu allen anderen Organen, namentlich dem Mastdarm-Plattenepithel." They³⁵ stated further as the cause of the disease as follows: "Früh eintretende kalte mit kalten Regengüssen andauernde Witterung, welche die in stets lebhafter Bewegung begriffenen Raupen zum Stillstehen bringt und ausserdem deren Nahrungsaufnahme herabsetzt oder auf Null reducirt."

Mr. J. M. KRASSILSCHTSCHIK³⁶ is inclined to believe that the presence of the so-called *Micrococcus lardarius* is characteristic of the Jaundice, while according to Dr. S. PROWAZEK,³⁷ the Jaundice is caused by the presence of *Chlamydozoan bombycis*, and on the polyhedral bodies he stated thus: "Die polyedrischen Körnchen fasse ich als spezifische Reaktionsprodukte der Wirtzellen auf das Virus auf."

33. J. BOLLE: Der Seidenbau in Japan. p. 121. 1898.

34. F. A. WACHTL und K. KORNAUTH: Mittheilungen aus dem Forstlichen Versuchstation Oesterreichs. p. 26—27.

35. loc. cit. p. 29.

36. J. M. KRASSILSCHTSCHIK: Sur les Microbes de la Flacherie et de la Grasserie des Vers a Soie. Compt. Rend. p. 428—429. 1896.

37. Archiv f. Protistenkunde. p. 361—363. 1907.

Messrs. A. CONTE and D. LEVRAT³⁸ have confirmed the cause of the Jaundice as follows:— "Ainsi donc, dans la grasserie, la cellule subit une dégénérescence tout à fait spéciale, caractérisée par la formation de cristaux intranucleaires;" but it seems to me that they do not discuss further the details of the process how and why the cells of the tissues or organs of the silkworms undergo degeneration.

As I have already stated before, the symptom of the Jaundice is not only the turbidity of the blood of the silkworms, which indicates certain advanced stages of the disease. In its earlier stages, the blood remains clear and transparent (in the case of yellow or green races, it is more or less of a yellowish color), and can not be distinguished from that of the healthy worms, but the polyhedral bodies of various sizes are almost always present in the cells, which form various tissues or organs particularly the fatty tissue and peritracheal membrane, silk-gland, hyperdermis &c.

Now if we harden the tissues or organs of the diseased worms with FRENZEL'S solution, and stain with carbol fuchsin, picric acid, hæmatoxylin, fuchsin &c., the polyhedral bodies readily take up the stains. In the cells of the diseased fatty tissues, hypodermal cells &c., we can always find the polyhedral bodies of various stages.

These polyhedral bodies are always produced within the nucleus of the cell, and not in the cytoplasm as already stated by several authors. In the nucleus of some cells, there can be seen certain numbers of minute dots of nearly equal sizes, but not in the cytoplasm. In other cells, the nucleus contains more or less larger dots or granules having more or less angular or polyhedral shape. These granules grow larger and larger within the nucleus, until the latter is nearly filled up with them. As the results of the growth of the granules, the latter assume an angular or many sided form, becoming polyhedral. Further the nucleus grows larger until it occupies the cell. In many cases, as a result of the abnormal growth of the nucleus, the membrane of the cell in which it is embedded ruptures, and the nucleus containing the granules becomes free

38. Laboratoire d'Etudes de la Soie. p. 58. 1906—1907.

and is thrown into the blood; or at the same time with the rupture of the cell membrane, the nuclear membrane breaks up too, and the granules contained in the nucleus are thrown out into the blood. Those sacs containing the polyhedral bodies usually floating in the blood of the diseased worms, which director J. BOLLE assumed as the cyst of *Microsporidium polyedricum*, appear to be nothing else than the nucleus containing the polyhedral bodies. Very often, these polyhedral bodies floating in the blood are imprisoned in the blood corpuscles, which look also like a sort of cyst.

The production of the polyhedral bodies within the cells of various tissues or organs of the diseased silkworms is still not ascertained, although there are several opinions among Japanese as well as Europeans. The opinions that the polyhedral bodies are the spores of *Microsporidium polyedricum* (Bolle), that they are produced as a "spezifische Reaktionsprodukte der Wirtzellen" by the presence of *Chlamydozoon bombycis* (S. PROWAZEK), or of *Micrococcus lardarius* (J. M. KRASSILSCHITSCHIK), or that they are produced as the result of degeneration of cells (A. CONTE and DE LEVERAT) are more or less correct; but none of them appears to me to point out distinctive cause of the production of the polyhedral bodies, i.e. of the Jaundice or Gelbsucht. According to my experiments performed during the past years, the production of the polyhedral bodies or of the Jaundice does not appear only to depend upon the presence of some parasitic micro-organisms, but upon several causes affecting the silkworms more or less.

As I have already stated before, the symptoms of the Jaundice of the silkworms are evidently not only the turbidity of their blood, which is due to the presence of the polyhedral bodies; but the presence of the polyhedral bodies in the organs or tissues of silkworms without regard to external symptoms as well as the presence of the same bodies in the blood, should be the main cause of the Jaundice. According to my opinion, the true or primary causes of the Jaundice of the silkworm are not simple, but multifold; but the results of the disease are, in all cases, the production of the polyhedral bodies as a secondary effect.

Those which I call the primary causes of the Jaundice as I have already discussed before, are:—

1st. The Jaundice is produced by the inoculation of the microbes, which are isolated from the milky fluid of the diseased worms, viz: the silkworms infested by these microbes, contain always the polyhedral bodies, as a secondary effect of these infestations.

2nd. When the silkworms are subcutaneously injected with the turbid milky fluid of the diseased worms, many of them become subject to Jaundice, as director J. BOLLE has experienced; on this fact, he concluded that the disease is produced by the contagion of the polyhedral bodies lying in the diseased worms, and thus he gave them the name *Microsporidium polyedricum*. But I can not agree with him, for according to my experiments, the production of the diseased worms (Jaundice) by the injection or inoculation of the milky fluid is not due to the introduction of the polyhedral bodies, but to that of the microbes, which are always present in the injected fluid.

3rd. The Jaundice of the silkworm is almost always produced by giving them food leaves bearing certain quantities of formalin. This teaches us that the action of formalin introduced into the body of the silkworms has the power to produce the polyhedral bodies characteristic of Jaundice.

The feeding of silkworms with the leaves of *Cudrania triloba*, *Taraxacum officinale* Wigg. var. *corniculatum* &c. more or less foreign to silkworms is considered as one of the causes of Jaundice.

4th. The interruption of the respiration of silkworms may also produce Jaundice, in this case, the external symptoms of the disease may or may not appear; but the contents of their body always contain the polyhedral bodies.

5th. The silkworms infested by maggots or suffering from flacherie and other diseases contain more frequently the polyhedral bodies without regard to the presence of the external symptoms or of the turbid blood characteristic of Jaundice.

6th. The polyhedral bodies readily take up certain coloring mat-

ters. They do not show any phenomenon characteristic of living organisms even after treatment with various culture media.

They are always produced within the nucleus of the cells forming the organs or tissues of the silkworms, as a result of certain irritation or action caused by microbes, parasitic maggots or by feeding the worms with the leaves of different plants excepting the mulberry, or by giving them food leaves bearing some chemical reagents.

7th. The hybrids or crosses of the wild ♀ and domestic ♂, as well as those of the domestic ♀ and wild ♂ silkworms are usually much affected by Jaundice, the latter appearing to be more liable to it than the former.

Finally, I conclude that the formation of the polyhedral bodies within the nucleus is undoubtedly to be ascribed to the degeneration or atrophy of the contents of the nucleus, due to more than one cause.

At the close of this work, I must express my heartiest thanks to Mr. S. KONNO, who has spent his whole time in assisting my present investigation during the last seven years.

August, 1909.

EXPLANATION OF FIGURES.

PLATE II.

Fig. 1.	Polyhedral bodies.	Zeiss, F. oc. 4.
Fig. 2.	Deformed polyhedral bodies.	Zeiss, F. oc. 4.
Fig. 3.	Streptococcus. a.	Zeiss, Apoch. 1.5, Com. oc. 18.
Fig. 4.	„ b.	„
Fig. 5.	„ c.	„
Fig. 6.	„ d.	„
Fig. 7.	Colony of Streptococcus a, on potato at 8 days after inoculation at the room temperature.	
Fig. 8.	Colony of Streptococcus b on potato.	
Fig. 9.	„ „ c „	
Fig. 10.	„ „ d „	

PLATE III.

Fig. 1.	Colonies of Streptococcus a, on Agar slant.			
Fig. 2.	„	„	b	„
Fig. 3.	„	„	c	„
Fig. 4.	„	„	d	„

PLATE IV.

Fig. 1.	Agar stab culture of Streptococcus a.			
Fig. 2.	„	„	„	b.
Fig. 3.	„	„	„	c.
Fig. 4.	„	„	„	d.

PLATE V.

Fig. 1.	Gelatine stab culture of Streptococcus a.			
Fig. 2.	„	„	„	b.
Fig. 3.	„	„	„	c.
Fig. 4.	„	„	„	d.

PLATE VI.

Fig. 1.	Milk culture of Streptococcus a.			
Fig. 2.	„	„	„	b.
Fig. 3.	„	„	„	c.
Fig. 4.	„	„	„	d.

PLATE VII.

- | | | |
|---------|--|------------------------------|
| Fig. 1. | Section of the diseased fatty tissue, showing various stages of polyhedral bodies. | Zeiss, Hom. Im. 1/12. oc. 2. |
| Fig. 2. | Section of the diseased hypodermis. | " |
| Fig. 3. | " " | " |
| Fig. 4. | " " | " |
| Fig. 5. | " the diseased malpighian vessel. | " |
| Fig. 6. | " the diseased silk gland. | " |
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On the Silk Fish-Line (Tegusu).

BY

Prof. C. Sasaki, *Rigakuhakushi*.

With Plate VIII—X.

The silk fish-lines, vulgarly called "Tegusu" by the Japanese, are largely employed by our fishermen in all parts of our islands. They are not prepared in our country, and a large amount of them is annually imported from southern China; but it is a very strange fact that till now no body knows exactly how and from what materials they are prepared.

In some Chinese as well as in Japanese works, some facts about these fish-lines are given, but they are almost all fragmentary, and therefore we have not been able to get a clear idea about them.

In an old Chinese work (Kanton Shingo, vol. XXIV), the following statements are made: "Some wild silkworms found in Yōkō feed upon the leaves of camphor trees and *Liquidambar formosana*. In April, when the worms become mature, they are dipped in vinegar and then filaments are taken from their body. They are about 7-8 feet long and have a golden yellow tinge."

The same facts are repeated in two other Chinese works "Kanton Tsushi" and "Kōshushi."

Mr. A. Ino wrote in his great work—Shobutsu Ruisan, about these fish-lines as follows:

"In the spring 1849, I asked a Chinese about the silk fish-line, then he replied. The silk fish-lines are prepared from insects in Hainan and Fukken in China. This insect appears in the months of April and May, and feeds upon *Liquidambar formosana*."

"In July and August, when the insect matures, the gut is taken out from its body, and after being soaked in vinegar, it is stretched out into long filaments and exposed in the air. When dried, it becomes the silk fish-line."

Mr. H. OBARA¹ states that the silk fish-lines are prepared from two convoluted threads lying within the body of the mature larva of *Caligula japonica*. The threads taken out of the larva, after being soaked in vinegar for some time, are stretched out, and when they are dried up in the air, they become silk fish-lines of about 5-6 feet in length.

In the circular² No. 103: second series, Mr. C. HANNEN reported as follows:—

"The camphor tree worm (shochu) is not a native of the mainland of this province (Foochow); but as special attention has been directed to the subject, I subjoin a few particulars."

"At certain seasons of the year the worms fall off the trees, and then only are they fit for use; they are immediately gathered, broken in two, dipped in vinegar, and have their intestines drawn out. The gut, miscalled silk by Chinese, is, after having been thoroughly dried in the air, used for fishing lines..... The best quality comes from Hainan, and small quantities used formerly to be imported in junks for sale to the Liuchiu islanders at the high price of about \$300 per picul. Tempted by this figure, a few piculs were, several years ago, collected and brought hither, overland, from the mountains of Kiangsi, but of such poor quality that it only realised \$60 per picul; so the experiment was never repeated. For some time the trade with this port has ceased entirely, the Liuchiuans supplying themselves viâ Japan. These worms

1. H. OBARA: Tōdōhitsu Vol. IV.

2. The circular is published by the order of the Inspector General of Customs in China, 1881. p. 138.

exist also, it is said, on the camphor trees in Formosa, but numbers are destroyed by birds and none are collected."

In the same circular p. 155, Mr. A. LAY reports to the Inspector General of Customs in Peking about the silk fish-line as follows:—"Besides the above-mentioned Raw Silk and Silk Piece Goods, Silk fish-lines are exported from Kiunchow. They are said to be made from large caterpillars found on a tree grown in the interior of the island, which my predecessor in office, judging from the specimen of the leaves he obtained, was considered *Liquidambar formosana*. The average annual production amounts to 120 piculs, of which the greater part is shipped away, and is ultimately sent to Europe."

Mr. Y. OSAKI has the following in his pamphlet.³ "In preparing silk fish-lines, the full grown larva of *Caligula japonica* are collected, and opening their body on the dorsal side two transparent soft threads are picked out and soaked in a strong vinegar to which is added a few bluish coloring matter. As soon as the soft threads change white in the vinegar, they are taken out, and stretched out to 6-9 feet. These threads, when dried, become strong fish-lines...."

In 1886, Mr. S. UYENO gives the followings in his work—Sina Bokyeki Jiten: "Gioshi or Shochushi (Silk fish-line) is a product of China, and is prepared from the so-called Shochu (camphor worms). When the worms mature, they are collected, and by cutting out their body, thread-like intestines are taken out and by making them firm and hard, silk fish-lines are prepared."

The following is taken from Tsusho Isan⁴ "The silk fish-lines imported into Japan from China are a product of South China particularly of the districts of Canton and Cansii. In the city of Canton, the fish-lines are called "Tensangyoshi."

"The principal localities where the lines are largely produced are the island Hainan and Rateishū, and the worms from which they are prepared, appear in the months of spring, and become mature in the

3. Y. OSAKI: Shōchu Yōho (Japanese) 1886.

4. Commercial Report (Japanese). No. 23. p. 12.

months of summer."

"Those which feed upon the leaves of camphor trees are said to be the best."

"The principal localities, where the lines are largely produced, are Shinkei and the neighbouring localities. In the same districts, the worms feed upon camphor leaves as in Canton district. When the worms grow mature, they are collected from the trees and then put in vinegar for about 30 minutes. As the worm's thread (silk gland?) solidifies, it is stretched out to 3-5 feet in length, and thus the silk fish-lines are prepared."

In a most recent Chinese publication,⁵ some account of the worms producing the silk fish-lines are given as follows:

"The worms are greenish in color, and are provided with spines all over the body. The newly hatched worms are as fine as hairs, and the birds are very fond of them. After 5 to 6 days, the worms bear about 30 spines on their body. When mature, they are about 3 Sun⁶ in length, and are as thick as a thumb. Their food plants are willows, pears, camphor trees, and *Liquidambar formosana*. The fish-lines are prepared from the worm three times in a year, viz: May, June and July. The full grown worms spin cocoons on the stems or branches of the food plants. The cocoons are very hard and compact. The worms dislike cold climate as well as north-west wind."

"After a heavy rain, when the sun shines brightly, a north-west wind blows, and sudden changes of temperature follow, then a large number of the worms will die. The worms which appear in the months of July and August, even when mature, do not descend from the food plants, and spin cocoons on branches. The imprisoned worms thus protected from the coldness of the winter, emerge as a moth in the following spring and lay eggs."

All the above statements are as imperfect and fragmentary that we

5. Tensan Zago.

6. a "Sun" equal 3 cm.

can not obtain a clear idea of this interesting and valuable worm, from which the silk fish-lines so largely employed by our fishing men are prepared; but no detailed study has been made up to the present.

In 1905, Mr. J. KATSUMATA went to Hainan island for the purpose of collecting mainly birds and insects. Before his departure from Japan, I told him there should be found the worms producing the silk fish-lines on camphor tree or *Liquidambar formosana* as their food plants, and ordered him to send me the worm as well as the food plants.

In July of the same year, Mr. KATSUMATA sent me some alcoholic specimens of the worms, number of fresh cocoons, and dried specimens of *Liquidambar formosana*.

The fresh cocoons were kept in a hot house in order to examine what sort of moth will come out. Fortunately, on the 11th of February 1906, 2 female moths emerged from the cocoon, and in the following six days, more female moths appeared in succession, and laid eggs in groups; but all these eggs were unfertilized and none hatched out, so that I could not study the larvae.

In order to study the habits and metamorphosis of this moth, the food plants as well as the method of preparing the silk fish-line from the larva, I made a journey in South China (particularly Cansii, Canton, Hainan &c.) in the spring months of the last year.

On this occasion, I could collect the cocoons, larvae of all stages of growth, and their food plants.

As the results of my study, I have determined that the worms producing the silk fish-line are the larvae of *Saturnia pyretorum* Westwood belonging to the family Saturnidae.

Saturnia Pyretorum Westwood.

This moth seems to have been described by WESTWOOD⁷, and Sir HAMPSON⁸ described it also more recently, and thinks that *Saturnia*

7. WESTWOOD: Orient Entomology.

8. Fauna of British India, Moth. Vol. I. p. 23.

cidosa Moore⁹ is a synonym of the present species.

HAMPSON's descriptions of *Sat. pyretorum* Westw. are cited as follows:—

“♂ differs from *lindia* in the base of the fore wing being mostly dark, the medial area whitish, the postmedial area heavily suffused with fuscous so as almost entirely to obliterate the dentate lines, a white submarginal line; outer margin fuscous, ocellus dark, with a white streak and ringed with yellow and black, two deep red subapical patches. Hind wing similar, the whitish medial area very broad, the waved lines entirely obsolete, an abolescent red subapical patch. ♀ with a large tuft of hairs at the end of abdomen.”

Mr. HAMPSON did not describe sufficiently the female moth, and I think it is not quite useless to mention some of the characters of the female and male moths which I could obtain in 1906 and 1909.

Female moth (Fig. 1, Pl. VIII). Length 3.6 cm. Expanse of wings 11 cm. Body thick, broad and clumsy. Head small, greyish, eyes black, antennae dull yellow, rather long and doubly serrated. Pro-thorax pale greyish yellow. The meso- and meta-thorax covered thickly with brownish black long hairs. Abdomen marked alternately with light greyish and dull yellowish grey transverse bands, while the terminal end of the abdomen is covered thickly with brownish black hairs. The basal area of the fore wing bears a nearly triangular brownish black patch. The scales on the wings are very variable as I could find more than nine different kinds.

The venation of the fore and hind wings of the present species is more or less different from that of its allied species, viz: the median vein is rather conspicuous and runs throughout the discal cell, but its proximal end disappears gradually towards the discal vein. The anal veins are three, of which the first is only distinct near the base of the wings and becomes gradually indistinct towards the periphery of the wings; the second is conspicuous throughout its length; the 3rd is much reduced

9. Transact. Entom. Soc. Vol. II. 3rd series 1364—1366. p. 423.

in size, and its distal end meets the 2nd close to the base of the same. Venation of the hind wing is more or less different from that of the fore and the discal cell is marked lengthwise with a somewhat distinct median vein. The brownish black hairs at the end of abdomen are all modified into long spindle-shaped scales, which are marked on their surface with parallel longitudinal striations.

Male moth: Length 3 cm. Expanse of wing 8 cm. Body smaller than in the female. Head small, greyish black, antennae yellow, long serrated. Pro-thorax light pinkish white; the meso-thorax greyish black, the meta-thorax greyish. Abdomen is rather pointed, greyish black, with a greyish white transverse marking along the posterior margin of each abdominal segment. The end of abdomen is covered with a small clustre of greyish hairs. The markings on the wings are nearly similar to those of the female.

Eggs: Eggs are usually laid in clustres on the trunk or branches of the food plants (Fig. 2. Pl. VIII). They are nearly oval and more or less depressed. The average size of ten eggs is:—

Length 1.5 mm.

Breadth 1.2 mm.

Thickness 0.45 mm.

They are milky white and marked with imperfect net-work markings. At one pole is a dark minute spot (micropyl), which is surrounded by radially arranged, nearly triangular markings, and eight short streaks. All over the surface of the eggs, there may be found large numbers of minute openings, which communicate with the interior of the egg by means of very fine canals running obliquely through the egg-shell.

The number of eggs laid in a single clustre or mass is 40-50, and the mass is usually covered up with the blackish scales grown at the posterior end of the abdomen of the mother moth.

Metamorphosis of *Saturnia pyretorum*.

In the provinces of Cansii and Canton, the moths are said to appear

in the months of January and February, and lay eggs which hatch out in a few days.

The larva becomes mature in the months of May and June, and spins the cocoon on the branches or trunks of the food plants. The larva changes into a pupa at the end of three weeks after spinning the cocoon, and the moth emerges in the winter months of the following year as mentioned before.

Larva: 1st stage (Figs. 3, 3,a. Pl. VIII). Length 7 mm. Breadth of head 0.95 mm. Body dark greyish, brown, sparsely covered with brownish white slightly serrated hairs, antennæ black. The thoracic shield dark brown. Ventral surface of the body dull greyish brown. Pectoral legs blackish, abdominal legs dull greyish brown. Spiracles brown. There is a single blackish hair wart on each subdorsal, supra-spiracular and infra-spiracular line of each body segment. A wart on the subdorsal line is long and stout and bears 6-8 brown hairs. On each of the supra-spiracular lines of the 2nd to the last body segment, there is a single black hair wart bearing 5-8 brownish hairs; 1st body segment destitute of hair warts. The hair wart on each infra-spiracular line is also black and provided with 10-12 greyish white hairs. The numbers of hairs on a wart of the different lines are as follows:—

	Inf. Spir.	Sup. Spir.	Sub. dors.	Sub. dors.	Sup. Spir.	Inf. Spir.
1st body segment.	12	0	8	7	0	12
2nd "	10	7	8	7	8	10
3rd "	10	7	8	7	6	10
4th "	10	6	6	6	6	10
5th "	10	6	6	6	6	10
6th "	10	6	6	6	6	10
7th "	10	6	6	6	6	10
8th "	10	6	6	6	6	10
9th "	10	6	6	6	5	10
10th "	10	6	6	6	6	10
11th "	10	6	6	6	6	10
12th "	0	8	8	8	8	0

2nd stage (Figs. 4, 4, a. Pl. VIII). Length 17 mm. Breadth of head 1.54 mm., blackish, sparsely covered with white hairs, antennæ

black. Body greenish yellow. The dorsal, supra-spiracular, spiracular lines as well as spiracles black. The 1st body segment (pro-thoracic segment) bears dorsally a blackish \perp shaped marking. The 12th body segment (9th abdominal segment) is provided dorsally with a blackish Y shaped marking. Ventral surface of the body is dull greyish yellow. Pectoral legs as well as the claws of the abdominal legs dark brown. The outer side of the 5th abdominal legs bears a nearly oval black patch. The subdorsal, supra-and infra-spiracular lines of each body segment excepting the 1st, 2nd and the 12th, are provided with a single orange yellow cylindrical process bearing setae and bristles. A process on the subdorsal and supra-spiracular lines bears 4-5 setae, and a long bristle.

3rd stage (Figs. 5, 5, a. Pl. VIII). Length 3.0 cm. Head 2 mm. broad, light green, and bears blackish markings of variable shapes. In some individuals, the two sides of the frontal plate are lined with black, and the parietal plates have each a long blackish marking of irregular outline, while in others the posterior corner of the frontal plate is provided with a triangular black marking, and the hinder as well as the outer half of each parietal plate tinged black. Body light greenish yellow with bright deep green sides. The first body segment bears dorsally three blackish spots—one central dot-like, and the other two lateral more or less elongated. The 12th body segment is provided dorsally with a Y-shaped black marking. Sometimes the three arms are separated by a central colorless portion. Ventral surface of the three anterior body segments (3 thoracic segments) colored deep black, that of the remaining body segments light greyish blue. Pectoral and abdominal legs light greyish blue. The outer side of the 5th abdominal legs bears a black marking. The median dorsal line of the first four body segments marked with a series of blackish dots, that of the remaining body segments with a blackish line, extending for nearly the anterior two thirds of each of these body segments. On the subdorsal line there runs a thick blackish broken line, and on the supra-and infra-spiracular lines, there runs also a fine blackish broken line. On the subdorsal, supra-and infra-spiracular lines on each body segment, lies an orange yellow cylindrical process bear-

ing whitish hairs and bristles. The process lying on the subdorsal line bears a single long white hair and 4-5 brownish bristles, that on the supra-spiracular line, a single white hair and 4-5 brownish bristles, and that on the infra-spiracular line, 3 long white hairs and 4-5 brownish bristles.

4th stage (Fig. 6, Pl. VIII). Length 36 mm. Head 3 mm. broad. The colorations as well as the markings are nearly similar with the 3rd stage.

5th stage (Fig. 7, Pl. VIII. Fig. 7, a. Pl. IX). Length 5,5 mm. Head 4 mm. broad, light greenish yellow, with 2 small blackish dots—one on each parietal plate. Sometimes, in addition to the blackish dots on the parietal plates, there is a small blackish marking at the posterior corner of the frontal plate, and still in other cases only the marking on the frontal plate is present. Body light greenish yellow with bright deep green sides as in the previous stage. The 1st body segment bears in this stage a large distinct yellow dorsal chitinous shield with two oval discs bearing several stiff hairs. This shield has, at each lateral edge two small blackish dots.

In the anterior half of the dorsal surface of the 12th segment, there are four blackish spots, and in the posterior half of the same, three blackish spots, of which the one at the posterior is larger than the rest. Ventral surface of the body light greyish yellow. Pectoral legs light greenish brown, abdominal legs light greenish yellow and bear an oval blackish marking on the surface. The deep green dorsal line of each body segment is marked with 3 blackish spots. Each subdorsal line of each body segment is provided with a cylindrical process bearing mostly 6 spines. The supra- and infra-spiracular lines are yellow, and each provided with a yellowish blunt process on each segment. The process on the supra-spiracular line bears 4 brownish spines and a long hair, and that on the infra-spiracular line, 6 brownish spines and 3 long hairs. Both the upper and lower edges of these two lines are marked with a variable number of blackish dots or streaks.

6th stage (Fig. 8, Pl. IX). Length 8 cm. Head 6 mm. broad, light greenish yellow. The blackish markings on the frontal and parietal plates have disappeared. Body light greenish yellow with a deep green

stigmatic line.

The pro-thoracic shield is light yellow and bears two oval discs with several stiff hairs. The number and position of the hairy processes lying on the subdorsal supra-and infra-spiracular lines are exactly same as in the previous stage. The bristle and hairs on the processes vary more or less on the different lines, viz:

Each process on the subdorsal line of the 2nd body segment (2nd thoracic segment) with 6 brown bristles and 2 long white hairs. Each process on the subdorsal line of the remaining body segments (3rd to 12th), with only 6 brown bristles.

Each process on the supra-spiracular line of the body segment (2nd to 12th) with 6 brown bristles and a long hair.

Each process on the infra-spiracular line of the body segments (2nd to 12th) with 6 brown bristles and 2-3 long hairs.

The markings on the dorsal surface of the 12th body segment (anal segment) have disappeared; but the blackish marking on the outer side of the 5th abdominal segment still persists.

7th stage (Fig. 9, Pl. IX). Length 9.5-10.0 cm. Breadth of head 8 mm. The coloration of the head and body is nearly similar as in the previous stage; but the dorsal side of the body is somewhat lighter in color. The blackish markings on the head are either present or not, while those associated with simple eyes remain. Pro-thoracic shield light yellow with two elongated oval discs bearing stiff hairs. Spiracular line is marked with deep green broad band lined along the lower edges with a blackish broken line. Ventral surface of body dull olive green. Spiracles oval and blackish. Pectoral legs light brownish green; abdominal legs dull green with 3 long blackish patches, and a dull brown marking close to their lower surface. The entire surface of the body is covered sparsely with short white hairs. The bristles and hairs on the processes vary in number on the different lines of the body, viz:

Each process on the subdorsal line of the 2nd body segment (2nd thoracic segment) bears 6 bristles and 2 long white hairs, and that on the same line of each of the remaining body segments, 6 dark brown bristles

and no hairs.

Each process on the supra-spiracular line bears mostly 5 brown bristles and a single long white hair, and that on the infra-spiracular line, mostly 6 brown bristles and 3 long white hairs. There are no markings on the dorsal surface of the 12th segment, but a deep brownish black, nearly triangular marking still persists on the outer side of the 5th abdominal legs.

In the mature worms, the body becomes more or less soft and shorter, the color becomes light orange yellow, and the lively greenish streaks on the body become less pronounced.

The mature worms descend from the food trees usually during the forenoon when the weather is warm and still, and ascend again in the afternoon till about 5 o'clock. In windy or rainy weather, they never descend, but always remain on the tree.

Silk glands (Fig. 10, Pl. IX). In the mature larvae, they are strongly developed and occupy the greater portion of the body cavity. Each gland is a very long convoluted tube, in which three parts may be distinguished (excretory, reservoir, and secretory). The excretory part, which forms the beginning of the gland is a very fine slender tube, colored faintly yellow. The reservoir is much thicker and longer than the excretory part and colored deep orange yellow. The terminal portion (secretory) of the gland is slightly shorter than the reservoir. The beginning of this part is of nearly equal thickness with the reservoir, but it gradually becomes thicker towards the somewhat pointed end. The secretory part is transparent and nearly colorless, and easily to be distinguished from the colored reservoir.

The length of these 3 parts of the silk glands is as follows:—

Excretory part	8.3 cm.
Reservoir...	27.0 "
Secretory part	24.0 "

Cocoon and chrysalis (Figs. 11, 12, 13, 13,a. Pl. IX). The cocoon is pretty large, spindle-shaped, thick and compact in texture. Its longer and shorter axes are respectively 4.8 cm. and 2.5 cm. It is

colored dark greyish brown and highly glossy in appearance. The broad end of the cocoon is open while the abruptly pointed end is closed. The opening at the broader end is surrounded on its outer surface by a more or less swollen broad band, whose free edges are provided with a series of thick stiff threads, the ends of which meet together so as to close up the opening.

Injuries to worms: The worms are fond of a warm climate and dislike cold. If the temperature falls below a certain limit, growth is retarded, and if it becomes too low, the worms will fall down abundantly on the ground. They also suffer injury from strong wind and heavy rain, both old and young ones.

The birds do not devour the older worms on accounts of the stiff hairs, which cover the body. So far as my observations go, the worms are generally free from insect parasites, such as attack more or less the larvae of other bombycid moths.

Thus the worms are comparatively free from enemies, and grow in a healthy condition, so that most of them appear to complete the metamorphosis and perpetuate the kind.

Method of raising Larva of *Saturnia pyretorum*.

The food plants of the larvae are said by the Chinese to be camphor tree (*Cinnamomum camphora* Nees), Fonshu (*Liquidambar formosana*), pear trees, willows &c., but during my journey in the South China, I could find the worms feeding only upon camphor trees and Fonshu, but not upon pear trees and willows.

In Canton and Cansii districts of South China, the food plants are mainly camphor trees, and Fonschu is rarely met with.

The camphor trees grow naturally or are cultivated by the natives along road sides, in the valleys among the hills lying not far from their dwellings, or in many cases around their houses.

The main purpose of cultivating the camphor trees is to procure timber, in which regular trade is carried on with Hongkong, and the

cultivation of the worms is a matter of secondary importance. Camphor is prepared in small quantities only in a few localities from the leaves, and not from the wood. The roots, branches and trunks, which are not utilized for timber, are generally used for fuels.

The camphor trees of the above mentioned districts are on an average not older than twenty to thirty years; and older ones are very rare, and are mostly met with in the neighbourhoods of shrines, temples &c. The largest tree which I measured was about 7-8 feet in diameter. Seedlings are raised as follows:—When the fruits are ripe, they are collected, and are sown; or in other cases sowing is dispensed with and wild seedlings are simply collected. In many cases, the seedlings are planted in suitable localities, leaving certain intervals between them.

In the beginning of April, the worms are mostly in the 2nd and 3rd stages, and it is therefore probable that they hatch out in the months of February and March. Concerning the preparation of the fish line, the natives told me as follows:

When the worms become mature in the month of May, the worms are collected. There are only two silk glands in the body. These are now soaked in vinegar of the best quality for about five to ten minutes, and are then taken out. Now one end of each silk gland is coiled up tightly near one end of a long bamboo piece, which is stuck into the muddy walls of houses. The other free end of the same gland is held between the thumb and a finger, and the whole gland is quickly stretched out either horizontally or vertically until it reaches the length of seven or eight feet, and dried up in the shade.

After drying completely, the filaments are removed from the bamboo pieces on which one end was coiled up. Large numbers of the filaments are formed into a bundle and soaked in water in a very large earthen ware, in which they are kept for two days. After they are washed cleanly by rubbing the bundle between the palms of both hands, they are taken out of the water and are dried up. The dried filaments are nearly transparent and very strong against pull, and are ready for use.

It is said that the worms fed on camphor leaves furnish lines of a

much superior quality to those fed on the leaves of *Liquidambar formosana*; but examination of the two sorts has failed to bring out any such difference.

In Hainan island, the worms feed exclusively on *Liquidambar formosana*, where it grows extensively at the central districts of the island; and no camphor trees are here met with.

The food plants are either of natural growth or cultivated nearly in every valley or around the dwelling houses. Very often the food plant forms a large extensive forest. The trees are not usually very large, and the average diameter of the trunk is two to three feet. Those having a larger diameter are mostly cut down for timber and fuel, so that the remaining trees are kept within the limits mentioned.

In the beginning of May, the worms become mature, and commence to spin cocoons, which are found between leaves bound together by filaments, or in the axils of the smaller branches or in cracks or crevices in the bark of the trunk. Sometimes the mature worms descend to the base of the trunk, where they crawl about for some time, and ascend again in search of a suitable position for spinning cocoons. It is generally believed by the natives, that this descent is due to the need of drinking water on the part of the mature worms, and that the need for water is a certain indication of maturity.

Method of preparing the Fish-Lines in Hainan Island.

Gathering of mature worms:—As before stated, the mature worms descending from the trunks are picked, put into bamboo basket or strong linen sacs, and carried home. Those that do not come down but remain out of reach high on the trees are captured by means of a small funnel shaped bamboo basket provided with a long bamboo handle of about seven or eight feet (Fig. 17. Pl. X). If any mature worm is found on a high branch, the funnel is held up by means of a long handle to where the worm is resting and moved slightly to and fro until the worm falls into it. Those worms that remain on the highest portion of the tree

where the funnel can not reach, are allowed to spin cocoons where they like. The moths which emerge from the cocoon in the winter or spring of the following year will give rise to the offsprings. On being carried to the house for preparing the fish-lines, the mature worms are put into a large bowl of earthen ware (of about four feet in height and 1.7 feet in diameter at the middle portion) sufficient to fill it to nearly one half of its capacity. Then water is poured in until the bowl is full. In this way, the living worms are soaked in water for 12-24 hours. At the end of this time, the worms are either very inactive or partly dead.

These inactive or dead worms* are now taken out of the water, and the ventral surface of the 4th to 6th abdominal segments is opened by means of the thumbs of both hands, and the two long silk glands are drawn out of the body. These glands are then soaked in vinegar (strong and of the best quality) for some minutes, until the glands assume a whitish appearance. Then the glands are taken out of the vinegar and transferred into pure water in which they are washed, and the yellow coverings or walls are completely removed by rubbing the glands between two fingers. About 60 glands are then bound by their excretory portions to the bifurcated end of a long bamboo stick (about one foot in length), so that the reservoirs as well as the secretory portions hang down from the stick. The stick is carried by women, girls or boys to their own houses, and thrust in deep enough into the higher portion of the outside of the mud wall of the houses. Then the free hanging ends of the glands are caught one by one between two fingers, and are pulled out straight until the gland elongates no more, when the free end is tied to a fine short bamboo piece, which is also thrust into the mud wall. In this way, the silk glands are kept in the air stretched between two bamboo sticks stuck into the wall until it dries up when it becomes firm and strong.

When the filaments are completely dried up, they are removed from the bamboo sticks, and formed into bundles of 50 to 60 filaments each.

* The worms from which the silk glands were removed, are put together in a large flattened iron pan, and after cooking with lard, are relished by the natives.

These bundles are in turn rolled up into rings (of about $7/10$ to $8/10$ of a foot in diameter) and a number of them is put into a large bowl filled up with pure water, and soaked for three or four days (Fig. 18, Pl. X).

At the end of this time, each bundle is rubbed in water between the palms of both hands until all the impurities and knobs lying on the surface of the filaments are washed away. When these are entirely removed, the bundles are taken out of the water and unfolded, and then hang in the shade. When the bundles are partly-dry, each one is bound separately throughout their entire length with a long flax thread (Fig. 19, Pl. X). When completely dry up, they are taken down and rolled up again into rings and sold by weight.

Sometimes, a long bamboo stick with a number of well washed silk-glands is tightly stuck into a mud wall, while the free end of each gland is twisted around one end of a smaller stick. Each gland is stretched out into a filament by rapidly pulling away the smaller stick and leaving it stuck into a large bundle of branches or leaves of certain plants laid on the ground (Figs. 15, 16, Pl. X). In this manner, the filaments are dried and become very compact and strong. In other case, a wooden rod is also employed for stretching out the gland. It is about 5-6 feet long and $2/10$ of an inch in diameter, and has shallow cuts at certain intervals, and a hole at one end. In this case, one end of the well washed gland is wound around the bifurcated end of a small bamboo stick, and the latter is pushed into the hole of the rod. The free end of the gland is then held between two fingers and pulled away horizontally until the gland has reached a suitable length. When the free end is fixed into one of the cuts, the filament is kept extended until dry (Fig. 14, Pl. X).

Of the fish-lines prepared in these ways, two sorts can be distinguished. One is nearly transparent and pretty glossy in appearance. Lines of this sort are prepared from those silk-glands whose colored envelopes have been entirely removed during preparation. The other sort is more or less dull yellow or dull orange yellow in color and not so glossy as the first. This results from the imperfect removal of the envelopes

during the preparation. The first sort is much superior in quality and higher in price.

The Rearing of the Larva of *Saturnia pyretorum* West. in Formosa Island.

From the cocoons of the moth, which I have introduced into Formosa island from South China last summer, the moths commenced to appear in the first part of January* 1909, and laid eggs within a week after their appearance.

In ten days after oviposition, the larvae began to hatch out from the eggs. The newly hatched larvae fed with camphor tree leaves matured after six molts, and from the 20th April they began to spin cocoons. The observations made on a single larva under culture by Dr. T. SHIRAKI are as follows:—

2	January, 1909.	Moths appeared.
6	"	"	They laid eggs.
16	"	"	Eggs commenced to hatch out.
16-30	"	"	Duration of the 1st stage of larva.
30	January-1 February, 1909...	...				" " " 1st moult.
1-14	Febr., 1909.	" " " 2nd stage
14-16	"	"	" " " 2nd moult.
16-23	"	"	" " " 3rd stage.
23-25	"	"	" " " 3rd moult.
25	Febr.-7 March, 1909...	" " " 4th stage.
7-9	March, 1909	" " " 4th moult.
9-18	"	"	" " " 5th stage.
18-21	"	"	" " " 5th moult.
21	March-3 April, 1909	" " " 6th stage.
3-6	April, 1909...	" " " 6th moult.
6-20	"	"	" " " 7th stage.
20	"	"	The larva matured and began to spin cocoon.

* The temperature in January varied from 50°—60° F.

June, 1909,

EXPLANATION OF FIGURES.

PLATE VIII.

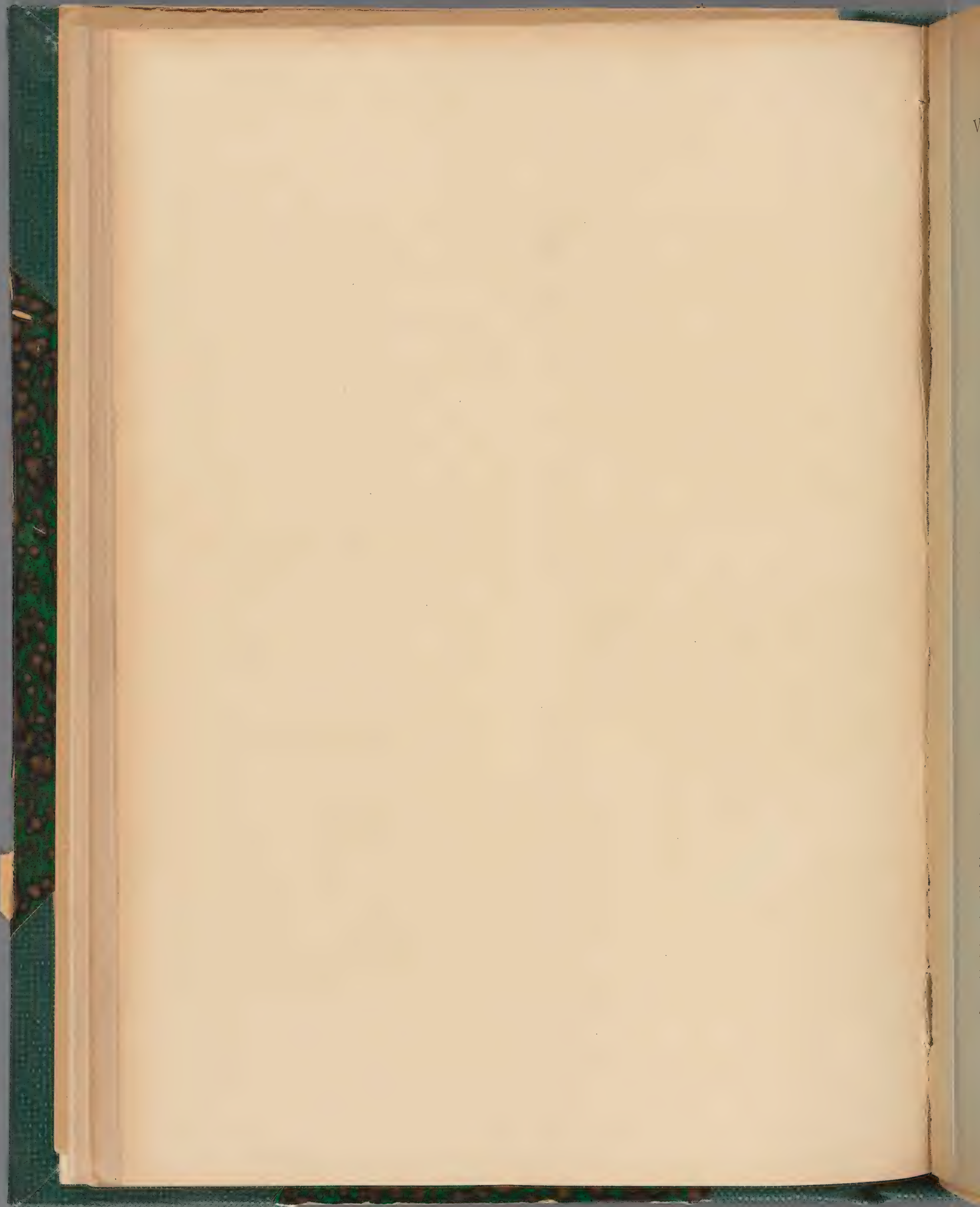
Fig. 1.	<i>Saturnia pyretorum</i> ♀.	Natural size.
Fig. 2.	Eggs clustre of ditto.	"
Fig. 3.	Larva of 1st stage.	Dorsal view (5/1).
Fig. 3, a.	" "	Lateral " (5/1).
Fig. 4.	" 2nd stage.	Dorsal " (2/1).
Fig. 4, a.	" "	Lateral " (2/1).
Fig. 5.	" 3rd stage.	Dorsal " (2/1).
Fig. 5, a.	" "	Lateral " (2/1).
Fig. 6.	" 4th stage.	Dorsal " (1/1).
Fig. 7.	" 5th stage.	Dorsal " (2/1).

PLATE IX.

Fig. 7, a.	Larva of 5th stage.	Lateral view (2/1).
Fig. 8.	" 6th stage.	" " (2/1).
Fig. 9.	" 7th stage.	" " (2/1).
Fig. 10.	Silk glands.	(2/1).
Fig. 11.	Cocoon.	(2/1).
Fig. 12.	Cocoon opened.	(2/1).
Fig. 13.	Pupa.	Dorsal view (2/1).
Fig. 13, a.	"	Lateral view (2/1).

PLATE X.

Fig. 14.	Wooden rod for preparing fish-line.
Fig. 15.	Mud wall with stretched fish-lines.
Fig. 16.	" "
Fig. 17.	Funnel with a long handle.
Fig. 18.	Rolled bundle of fish-lines.
Fig. 19.	Unfolded bundle of the same dried in air.



Vol. II. No. 2, PUBLISHED FEBRUARY 12th, 1910.

Price in Tokyo Yen 0.70.

THIS JOURNAL IS ON SALE AT

Z. P. Maruya & Co., L'td.

Tori Sanhome, Nihonbashi, Tokyo.

明治四十三年二月十日印刷
明治四十三年二月十二日發行

編纂兼發行者 東京帝國大學

印刷者

西谷弟松

東京市麻布區市兵衛町二丁目十六番地

印刷所

シヤパン、タイムス社

東京市麴町區內幸町一丁目五番地

賣捌所

丸善株式會社書店

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13,851

Vol. II. No. 3.

東京帝國大學
農科大學紀要

第二卷第三號

JOURNAL

(FORMERLY BULLETIN)

OF THE

COLLEGE OF AGRICULTURE,

IMPERIAL UNIVERSITY OF TOKYO.

TOKYO.

Published by the University.

APRIL 30TH, MEIJI XLIII (1910).

PUBLISHING COMMITTEE.

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A Further Contribution towards the Knowledge of the Panorpidae of Japan.

BY

T. Miyake.

With Plate XI and one Figure in the Text.

In my former paper, I published a list of the Panorpidae of Japan together with descriptions of ten new species.¹ The descriptions had been finished in October, 1907, and were immediately prepared for printing. The publication was, however, delayed owing to various circumstances, and was scarcely under way in September, 1908. In the meantime, I heard that Mr. NAVAS of Spain has published an essay on Neuroptera, describing a certain new species of Panorpidae from Japan.² I therefore asked the author at once for a copy of the paper, intending to make some further additions to my list of Panorpidae and also to ascertain if his species and my own may not be the same ones, so as to avoid as far as possible an unnecessary duplication of names. Unfortunately however his paper did not reach me before the publication of my paper, so that neither additions nor alterations could be made. Mr. NAVAS' work, a copy of which I owe to his kindness, contains descriptions of four new species of Panorpidae from Japan, namely:—

Panorpa Bourieri n. sp., Mem. Real. Acad. Cien. Art. Bar. Vol.
VI, no. 25, p. 20, (1908).

1. MIYAKE: A list of Panorpidae of Japan, with descriptions of ten new species, Bull. Coll. Agr. Tokyo Imp. Univ. vol. viii, No. 1 (1908).

2. NAVAS: Neuropteros nuevos, Mem. d. l. Real. Acad. de Cien. y. Art. d. Bar., vol. vi, No. 25 (1908).

Panorpa nipponensis n. sp., l. c.

Panorpa Drouarti n. sp., l. c. p. 21.

Panorpa dyscola n. sp., l. c. p. 22.

Adding them to my own list, the number of the Panorpidae of Japan now amounts to 30 species, of which 25 belong to the genus *Panorpa*, 2 to *Leptopanorpa*, 2 to *Panorpodes* and 1 to *Bittacus*.

Nevertheless it is necessary to critically compare NAVAS' species and my own. Though my paper was sent to the printer in October, 1907, and published in 1908, in the same year as that of NAVAS, if his paper was issued earlier in the year, the right of priority should belong to him. Now *P. nipponensis* of NAVAS and *P. brachypennis* of mine are closely allied, if not exactly alike. The principal differences between the two species are:—in *nipponensis* the wings are hyaline, in *brachypennis* ochraceous; in *nipponensis* "venulis in medio apicali albidis," in *brachypennis* they are fuscous. Moreover the wings of the latter are remarkably broad, probably broader in proportion than in any other species, so that this point appears to me to need further consideration before deciding on the possible synonymy of the two species. Now I have captured four female specimens on Usuitōge in Aug. 1908, among which there was one in which the transverse veins of the apical half were partly hyaline, and the broadness of the wings was less conspicuous than in the typical form. This leads me to consider the *nipponensis* of NAVAS and the *brachypennis* of mine to be one and same species, and I relegate my *brachypennis* to the list of synonyms.

Of *P. Drouarti* of NAVAS I have nothing to say.

Of *P. dyscola* of NAVAS, though it is very closely allied to *P. pulchra* of mine, I am still inclined to think that they are distinct species, because I have since captured a male of *pulchra*, which has no great difference in the wing markings as in the female, while *dyscola* has different wing markings in the two sexes. The details of the relationship of allied species *japonica*, *pulchra*, *dyscola* and *irregularis* will be discussed under the heading of *irregularis*.

It is possible that the *P. Bouvieri* of NAVAS is simply a variational

form of *P. Pryeri* M'L. This species is very variable, and one form was described in my former paper as var. *major*. The wing markings are especially variable, so that one is often misled to think that he has another species before him. But in the essential structural characters, the species is rather fixed, and deviations can usually be traced to the typical form without difficulty.

Thanks to new additions to my collection of Panorpidae, I have obtained direct knowledge of almost all the species known to occur in Japan. Only *P. macrogaster* of M'LACHLAN, two species of *Leptopanorpa* of M'LACHLAN (*L. Ritzemae* and *L. Sieboldi*) and the new species of NAVAS above mentioned are still unknown to me. There are besides 8 species, all of which I consider to be new to science, and are described in the following pages. About *P. irregularis* n. sp. only, I have slight doubt whether to consider it a distinct species or not. At present I am more inclined to think it distinct, and it is accordingly described in this paper.

Among the fine series of specimens of *P. japonica* I have discovered some forms closely allied to *P. macrogaster* M'L., and if the species nearly allied to *P. japonica* i.e. *P. pulchra*, *P. irregularis* and *P. dyscola* should happen to prove a single species—*P. macrogaster* should also be included in it without any hesitation.

Prof. MATSUMURA of Sapporo described *P. communis* L. in his 'Senchuzukai' (Thousand Insects of Japan), vol. 1, p. 164, pl. XI, fig. 6 (1904). It is a common insect in Europe having *the subcosta in the anterior wings reaching to the pterostigma*. As all the Japanese species hitherto known were exclusively characterized by the peculiarity mentioned by M'LACHLAN that "the subcosta in all the wings scarcely extends beyond the middle of costal margin," I have hesitated to count this exceptional form as belonging our fauna. Last year however I went to the Entomological Laboratory of Prof. MATSUMURA of Sapporo and saw his *communis* in his cabinet. There was no doubt that it was a typical *communis* with the above-stated character. And as it was captured in series at the Lake Towada in the Province of Akita, a place surrounded

by a range of high mountains, I think I can not but recognize it as one of our faunistic species, and at the same time the above mentioned peculiarity of our fauna is set at naught.

I have discovered a second species which bears the last mentioned character but which has no other resemblance with *communis*. It is to be considered as a new species which is described beyond in this paper as *P. gokaensis*. It was captured by me at Gokanoshō in Kiushu, a locality surrounded by many folds of mountain ranges, and very difficult of communication even at present. Gokanoshō proved a very good place for collecting *Panorpa* as not only the species just mentioned but a number of interesting ones were obtained there.

After all it is a very interesting fact that a character common in European formes proves to be a rare and exceptional one in the Japanese fauna and conversely the peculiarity of our fauna appears to be rare and exceptional in the European formes. Indeed *P. alpina* of Europe is the representative of this peculiarity.

Panorpa appears in Japan from the end of April to the end of September, and May is the best time for collecting.

I have to mention all the species of our Panorpidae known up to the present together with new species described in this paper. (Italics denote synonyms and * doubtful species).

- | | |
|--|-------------------------------------|
| 1. Panorpa † japonica Thunb. | (<i>Shiriagemushi</i>). |
| 2. P. macrogaster M'Lach.* | (<i>Ō-shiriagemushi</i>). |
| 3. P. pulchra Miyake. | (<i>Aya-shiriagemushi</i>). |
| 4. P. dyscola Navas. | (<i>Kurofu-shiriagemushi</i>). |
| 5. P. irregularis n. sp.* | (<i>Midare-shiriagemushi</i>). |
| 6. P. sinanoensis Miyake. | (<i>Usumon-shiriagemushi</i>). |
| 7. P. nihonensis Miyake. | (<i>Ko-shiriagemushi</i>). |
| 8. P. rectifasciata Miyake. | (<i>Obi-shiriagemushi</i>). |
| 9. P. obscura n. sp. | (<i>Ko-obi-shiriagemushi</i>). |
| 10. P. chuzenjiensis n. sp. | (<i>Tsumaguro-shiriagemushi</i>). |
| 11. P. ochraceopennis n. sp. | (<i>Kibane-shiriagemushi</i>). |
| 12. P. trizonata Miyake. | (<i>Misuji-shiriagemushi</i>). |
| 13. P. ochracea Miyake. | (<i>Kihada-shiriagemushi</i>). |
| 14. P. Klugi M'Lach. | (<i>Bekkō-shiriagemushi</i>). |

† See the foot-note on page 187.

- | | | | |
|-----|--------------|------------------------------|--|
| 15. | P. | Drouarti Navas. | (<i>Iime-shiriagemushi</i>). |
| 16. | P. | nipponensis Navas. | (<i>Maruhane-shiriagemushi</i>). |
| | P. | <i>brachypennis</i> Miyake. | |
| 17. | P. | Pryeri M'Lach. | (<i>Pryer-shiriagemushi</i>). |
| | P. | <i>Bouvieri</i> Navas. | |
| 18. | P. | leucoptera Uhl. | (<i>Futasuji-shiriagemushi</i>). |
| 19. | P. | Wormaldi M'Lach. | (<i>Kiashi-shiriagemushi</i>). |
| 20. | P. | multifasciaria n. sp. | (<i>Hoso-madara-shiriagemushi</i>). |
| 21. | P. | striata Miyake. | (<i>Suji-shiriagemushi</i>). |
| 22. | P. | Lewisi M'Lach. | (<i>Hosoba-shiriagemushi</i>). |
| 23. | P. | bicornuata M'Lach. | (<i>Momoguro-shiriagemushi</i>). |
| 24. | P. | nikkoensis Miyake. | (<i>Nikkō-shiriagemushi</i>). |
| 25. | P. | Takenouchii Miyake. | (<i>Hoshi-shiriagemushi</i>). |
| 26. | P. | cornigera M'Lach. | (<i>Hoso-obi-shiriagemushi</i>). |
| 27. | P. | magnicauda n. sp. | (<i>O-hasami-shiriagemushi</i>). |
| 28. | P. | gokaensis n. sp. | (<i>Maye-futasuji-shiriagemushi</i>). |
| 29. | P. | communis L. | (<i>Madara-shiriagemushi</i>). |
| 30. | Leptopanorpa | Ritzemae M'Lach. | (<i>Hoso-shiriagemushi</i>). |
| 31. | L. | Sieboldi M'Lach. | (<i>Tsumaguro-shiriagemushi</i>). |
| 32. | Panorpodes | paradoxa M'Lach. | (<i>Sukashi-shiriagemushi-modoki</i>). |
| 33. | P. | singularis n. sp. | (<i>Kasuri-shiriagemushi-modoki</i>). |
| 34. | P. | decorata M'Lach. | (<i>Matamon-shiriagemushi-modoki</i>). |
| 35. | P. | apicalis n. sp. | (<i>Tsumaguro-shiriagemushi-modoki</i>). |
| 36. | Bittacus | sinensis Walk. | (<i>Kaganbo-modoki</i>). |

A synopsis of these species is given below, the species of Genera *Leptopanorpa*, *Panorpodes* and *Bittacus* being omitted.

A. Subcosta in the anterior wings extending to the pterostigma.

gokaensis n. sp.

B.* Subcosta in all the wings extending to the costal margin scarcely beyond its middle.

a. Apex of wings with broad dark space.

a¹. Both wings with only a small pterostigmatical patch; the broad (pterostigmatical) fascia entirely absent.

chuzenjiensis.

* Just as I was sending the last proof of this paper to the printer, I came across the paper of Dr. G. Enderlein in Zool. Anz. Bd, XXXV, Nr. 12/13 (Feb. 1910), in which he proposes a new genus *Aulops* for this group.

b¹. Both wings with only one broad (pterostigmatical) fascia connecting costal and hind margins.

a². Apex of wings elliptical, wings moderately broad.

a³. Wings hyaline.

a⁴. Fascia furcate externally, or with irregular edges.

a⁵. No spots before the fascia, or some spots before the fascia present.

a⁶. Fascia furcate, spots absent or a series of spots before the fascia may be present, appendages straight and not much divalicate.

japonica.

b⁶. Fascia furcate, two series of spots before the fascia present, appendages straight and slightly more divalicate than the preceding.

irregularis n. sp.

c⁶. Fascia not furcate, appendages straight and much divalicate.

niphonensis.

d⁶. Fascia not furcate, appendages slightly curved.

sinanoensis.

b⁵. A narrow oblique streak before the fascia present.

a⁶. Another streak present, so as it forms with the other streak V-shaped markings.

pulchra.

b⁶. With one oblique streak.

a⁷. Apical dark portion furcate at middle; female unknown.

Drouarti.

b⁷. Apical dark portion evenly curved.

dyscola.

b⁴. Fascia with sharply defined edges, no spot present.

rectifasciata.

b³. Wings tinged with ochraceous or testaceous.

a⁴. Pterostigmatical fascia broad, its inner margin furcate or connected with some spots.

ochraceopennis n. sp.

b⁴. Pterostigmatical fascia moderate, or rather narrow, with sharply defined edges.

a⁵. Fascia moderate; cheliferous appendages V-shaped; size small.

obscura n. sp.

b⁵. Fascia rather narrow; cheliferous appendages slightly curved; size large.

ochracea.

c⁵. Fascia rather narrow; cheliferous appendages much curved; size small.

Klugi.

b². Apex of wings rounded; wings rather broad.

brachypennis (= *nipponensis*?).

c¹. Both wings with two broad fasciae.

trizonata.

b. Apex of wings with narrow dark space or dark space absent.

a¹. Wings with the complete pterostigmatical fascia.

a². Markings of wings consisting essentially in streaks or some curved lines.

a³. Wings with many streaks (usually 3) between the apex and the fascia.

a⁴. Chelae as usual.

a⁵. Markings not deeply coloured; appendages rather broad, long, with pointed apex.

Wormaldi.

b⁵. Markings deeply coloured; appendages rather broad, short, with rounded apex.

striata.

b⁴. Chelae much dilated at the basal portion.

multifasciaria n. sp.

b³. Wings with some spots or an irregular streak consisting in connected spots between the apex and the fascia.

Pryeri.

c³. Wings with a lunate spot between the apex and the fascia.

cornigera.

- d³. Wings with no spot between the apex and the fascia.
magnicauda n. sp.
- b². Markings of wings consisting essentially in spots.
Takenouchii.
- b¹. Wings with the incomplete pterostigmatical fascia.
- a². Three spots at the costa before the pterostigma.
leucoptera.
- b². Two spots before the pterostigma.
nikkoensis.
- c². One spot before the pterostigma.
bicornuata.
- d². No spot before the pterostigma.
Lewisi.

In the following pages 7 new species of the genus *Panorpa*, two new species of the genus *Panorpodes* and the male of *Panorpa Wormaldi* are described. The male of *P. Wormaldi* is described here for the first time in my knowledge.

1. ***Panorpa** *ochraceopennis*** n. sp. (*Kibane-shiriagemushi*).

(Pl. XI. figs. 1, 1a, 1b, ♂.)

Body blackish piceous; rostrum black; antennæ and palpi fuscous to piceous; legs ochraceous or fuscous ochraceous.

Wings moderate, tinged with ochraceous yellow; a little narrower compared with that of *P. japonica*; apex elliptical; a broad blackish fascia beyond the middle, the outer edge of which is almost always sharply defined, though in a few cases it is slightly furcate, and the inner edge always furcate on its upper and lower portions; the upper branch arises just beyond the middle running to the costal margin oblique in a direction contrary to that of the fascia; when it is produced to the costal margin it encloses a round untinged spaces between the branch and the fascia; the lower branch runs always along the hind margin and usually more prominent than the former; both branches especially the lower one en-

* See the foot-note on page 187.

larges frequently into an irregular patch, which when very pronounced may become continuous with each other; the furcation of the inner edge predominates usually on the fore wing, the inner edge in the hind wing in many examples defined sharply like the outer one, seldom however it is furcate or the furcate portions reduced to marginal spots; apex also broadly blackish with almost straight or slightly sinuated internal edge; longitudinal and transverse veins of the basal half and the portion where they cross the markings mostly black; those of the space between the fascia and the apical portion yellowish.

Abdomen blackish or blackish piceous; in the ♂ the posterior margin of the 3rd dorsal segment is produced into a short broad median lobe, which is however shorter and broader than that of *P. japonica* or its allies, and therefore less conspicuous than in the latter species; 6th, 7th and 8th segments almost equal in length though they are very slightly longer in progression; 6th segment very stout and obconical, while 7th is very slender and cylindrical and much like the 8th, which is also very slender, so that the abdomen is abruptly attenuated from the 7th towards the extremity; 9th (cheliferous segment) rather smaller and less stout than in *P. japonica* and its allies; chelae long, the appendages rather long and moderately curved as in *P. Klugi* and the distal half bent downwards between the two lateral pieces.

Expanse 29 mm.—27 mm.

Three males and a female obtained on Mt. Nasu, Tochigi-ken, July 19, '08; a male and a female at Hibara, Yamagata-ken, July 24, '08; a male on Mt. Yudono, Yamagata-ken, July 27, '08; a female on Mt. Usui, Nagano-ken, Aug. 3, '08; by Mr. Habutsu of the Agricultural College and by the author; four female specimens in the collection of the College obtained at Nikko. Aug. 31. 1895.

This species is decidedly different from other species in having an ochraceous tinge on the wings, the inner edge of the fascia of which being irregularly furcate and the abdomen being abruptly attenuated from the 7th segment towards the end, the cheliferous appendages of which are also somewhat peculiar to the species. The ochraceous tinge of the wings

varies according to degree of maturity as the freshly emerged insects have usually a very pale coloration. There is a male in the collection, in which the wing is colourless as in *P. japonica* with very pale markings and fuscous abdomen.

The distribution of this species might, so far as I know, be restricted to a comparatively very small area, as all the specimens before me were captured in the north-east middle part of Honto and I have never received any example from the south west part of Honto or from Shikoku and Kiushu.

2. *Panorpa magnicauda* n. sp. (*Ō-hasami-shiriagemushi*).

(Pl. XI. figs. 6, 6a, 6b, ♂.)

Body black; rostrum black; antennae and palpi piceous; legs yellow with very slight fuscous tinge; extremity of each joint and terminal joint of tarsi fuscous with the claws testaceous.

Wings hyaline, moderate, with the apex elliptical; a rather narrow blackish oblique fascia beyond the middle (about two thirds from base of wing), which is somewhat attenuate towards the posterior margin; a small blackish apical patch (the patch encloses a small pale spot in the fore wing); basal half of costal margins of fore and hind wings and posterior margin of fore wing suffused with blackish; a very small blackish point, almost insignificant in the posterior wing, at a place one third from the base, rather nearing the posterior margin; longitudinal and transverse veins mostly piceous.

Abdomen black, very peculiar in shape; 1st segment just as in other species; 2nd and 3rd segments very short and almost equal in length; in the specimen (male), the 3rd dorsal segment is produced in its middle into a short but broad lobe; 4th, 5th and 6th cylindrical with testaceous pleural membranes; of the three segments the 6th is longest and the 4th shortest; 7th cylindrical, obliquely truncate anteriorly from dorsal side posteriorly to ventral side; the ventral apex of 7th segment attenuate and pointed on each side so as to form two conspicuous spines (without a

parallel among the hitherto examined species); 8th rather short, scarcely longer than 6th and 7th segments, conical and much narrower than the other segments; 9th segment very large with much elongated lateral pieces; chelae rather short, slightly incurved, with the testaceous apex; cheliferous appendages very conspicuous, elongated, piceous, and extending over the middle of chelae; the distal end of the appendages is overlapped the left piece up, so as to show an O-shaped structure.

Expanse 31 mm.

A single male specimen obtained at Goka-no-shō, Kiushu, by the author, on May 27, 1908, in the collection of the Imperial Central Agricultural Experiment Station.

This species is allied to a certain extent to the *P. Davidi* of NAVAS, which, he says, is similar to *P. cognatae* Ramb., so far as the wing markings only are concerned. But in the structural characters, especially in the form of cheliferous segments, the present species has no common points with the other two. It is surely a distinct species and moreover an unparalled form.

3. *Panorpa gokaensis* n. sp. (*Maye-futasuji-shiriagemushi*).

(Pl. XI. figs. 3, 3a, 3b, ♂.)

Body totally black; rostrum, antennae and palpi black; legs fuscous yellow, with the extremity of each joint and some terminal tarsi fuscous; claws testaceous.

Wings hyaline, with the apex elliptical; a rather narrow (compared with that of *P. japonica*, etc.) blackish postmedial fascia from the pterostigma to the posterior margin oblique in posteriorly inward direction; in the fore wing a likewise narrow blackish antemedial fascia is present, slightly oblique in direction contrary to that of the postmedial one, that is in posteriorly outward direction; in the female specimen the fascia is extended from the costal margin backwards, in the male terminating before reaching the posterior margin; a rather small blackish apical

patch, with well defined inner edge; the subcosta of fore wing extending to the pterostigma; costa, subcosta and radial veins black; the remaining longitudinal veins and transverse veins mostly fuscous yellow except where they cross the fascia, where they are black.

Abdomen rather short, black; in the ♂ the posterior margin of the 3rd dorsal segment is not produced into a lobe (unlike any other of our *Panorpa*); 6th cylindrical; 7th narrower and longer than 6th and almost equal to the 8th; 9th rather smaller but stouter than in *P. japonica*; lateral pieces very stout and rounded; chelae shorter than the segment, almost straight, being very slightly curved towards the extremity which is testaceous; appendages rather long, slightly curved, approximating at the base and apex.

Expanse, ♂, 30 mm; ♀, 33 mm.

A male and a female specimen captured at Goka-no-shō in Kiushu, by the author on May 27, 1908.

This species shows a remarkable and peculiar feature, unlike all other species of our *Panorpa*, that is, sub-costa of the anterior wing reaches to the pterostigma as in most European species, the words "subcosta in all the wings scarcely extends beyond the middle of the costal margin," not holding in this species. Besides, the dorsal side of the 3rd abdominal segment of male in this species is not produced into a median lobe, as in other species of our *Panorpa*. In these peculiarities the present species is undoubtedly allied to some European forms, especially to *P. communis* L., *P. connexa* M'L. and *P. annexa* Selys. From those species however it is certainly different in the wing markings and the structure of the cheliferous segment, although the form of the appendages shows a certain common feature with the three species. So far as the wing markings go, the species is allied to *P. magnicauda* mihi and *P. Davidi* of NAVAS. In *magnicauda* the subcosta is not extended to the pterostigma as in the present species, and cheliferous segment is very peculiarly constructed. *P. Davidi*, which is according to NAVAS nearly allied to *P. cognatae* Ramb, and has similar wing markings as the present species, differs from it in many other respects, and moreover though NAVAS does not mention the

3rd abdominal segment, that of *cognatae* is "considerably produced in the middle," according to M'LACHLAN'S statement. Anyhow it may safely be said that the present species is in some points of importance allied to the European form, a fact really worth mentioning on account of the isolated character of the locality where it occurs, viz. Goko-no-shō in the interior of Kiushu, surrounded by many ranges of mountains. I have found the species not uncommon there, occurring side by side with a number of other Panorpids.

4. **Panorpa obscura** n. sp. (*Ko-obi-shiriagemushi*).

(Pl. XI. figs. 2, 2a, 2b, ♂.)

Body blackish piceous; rostrum blackish piceous, with the palpi testaceous; antennae and eyes testaceous; legs yellowish.

Wings moderate, slightly tinged with ochraceous yellow; with the apex elliptical; a broad (but narrower compared with that of *P. japonica* Thunb. or *P. rectifasciata* Miyake) fuscous fascia beyond the middle with slightly wavy inner and outer edges; apex also fuscous, with the inner edge slightly sinuous beyond middle; no points or patches otherwise present; longitudinal and transverse veins fuscous, of which the latter is somewhat lighter in colour.

Abdomen blackish or blackish testaceous; in the ♂ the posterior margin of the 3rd dorsal segment of the abdomen is produced into a short broad median lobe; 6th segment stout, cylindrical; 7th slightly narrow and longer than the 6th; 8th slightly longer and much narrower than 7th; 9th rather slender, the chelae ochraceous, slender, the appendages rather long, divaricate towards its extremity, so as to show V-shaped structure.

Expanse 27 mm-32 mm.

Two males and a female captured by the author on Hibara-tōge, Yamagata-ken, on July 24, '08 and two female specimens in the collection of Agricultural College, captured by Prof. SASAKI at Ichinohe and Sambongi, Aomori-ken, Aug. 15, 29, '02.

This species is allied to *P. rectifasciata* Miyake in its wing markings, but as stated above, the ground colour of its wings is ochraceous yellow, while that of *rectifasciata* is almost hyaline, the post medial band of the former is wavy towards the ends and much narrower in proportion, while that of the latter is always straight, with very sharp edges and much broader than in the former. Moreover the structure of the cheliferous appendages readily separates the two species, because in the present species each portion of the appendages is almost straight and divaricate distally so as to show a V-form while that of *rectifasciata* is much shorter and more curved, each portion running somewhat close to each other. This species seems to be very rare as it occurs in a small restricted place.

5. ***Panorpa multifasciaria*** n. sp. (*Hoso-madara-Shiriagemushi*).

(Pl. XI. fig. 5 ♀; fig. 5a apex of abdomen of ♂; 5b Chelae).

Body blackish piceous; rostrum black above, the lateral and under sides with the palpi ochraceous yellow; antennae blackish piceous; legs ochraceous or fuscous yellow.

Wings hyaline, with rather acuminate apex; black or rarely fuscous markings as follows:—in some specimens there is a streak between the subcostal and radial veins running from the base to the end of the former; in some specimens this streak is reduced to a very short one situated near the end of subcosta; and again in some specimens this streak is entirely absent; two conjoined (rarely separate) elongated spots situated transversely just at end of the subcosta; an irregular oblique fascia commencing at the anterior margin beyond the middle (at pterostigma) and ending posteriorly just at middle of the posterior margin; posterior margin with an irregular streak from base to the preceding fascia, the inner half running along the margin and the outer half consisting of two conjoined spots running a little anteriorly apart from the margin, so that it includes two small quadrate hyaline patches between them; over the middle of the oblique fascia, outwardly and posteriorly starts a fascia ending at the posterior margin; an irregular, sometimes discontinuous,

fascia from the pterostigma (therefore uniting with the above-mentioned oblique fascia) to the posterior margin; another irregular fascia beyond the one just mentioned from the anterior to the posterior margins, furcate in its lower half or broken into short streaks; a curved streak just before the apex, in some specimens however this is divided into many small spots; a rather indistinct short streak between each of the longitudinal veins along the apex; longitudinal veins, except those of median and basal parts of wing, black; the rest and most of transverse veins black; wings somewhat iridescent.

Abdomen rather short, piceous, in the single male specimen, the ventral side and the cheliferous segment are ochraceous, (I cannot determine whether this is due to the degree of maturity, as the specimen is in an immature condition) while in the female specimens the abdomen is tinged piceous all over; posterior margin of the 3rd dorsal segment produced into a short but broad median lobe; 6th segment broadest of all the segments just as in the somewhat allied *P. striata* Miyake; 6th and 7th of nearly equal length; 8th longer than the 7th; cheliferous segment short but rather large, ochraceous; lateral pieces strongly rounded; chelae very short, but the basal $2/3$ much dilated; cheliferous appendages not satisfactorily recognizable on account of shrinkage.

Expanse, ♂, 28 mm; ♀ 30 mm-32 mm.

A male captured by Baron TAKACHINO on Hikosan, Kiushu, on April 28, '02; a female specimen in the collection of the Agricultural College, captured in Gifu by Mr. TERADA on May 29, '09; a female in the collection of the Imperial Central Agricultural Experiment Station captured at Goka-no-shō, Kiushiu, by the author on May 27, '08; a female in the collection of the Nawa Entomological Laboratory obtained at Kasugamura, Gifu-ken, May 16, '03.

This species is closely allied to *P. Wormaldi* M'L. and *P. striata* Miyake in the wing markings, though it can readily be distinguished by the form of the cheliferous appendages of the male. Although these three species have closely similar wing markings, each species has some peculiar features of its own. In all, the posteriorly furcate pterostigmatical fas-

cia, with the inwardly situated anterior and posterior marginal fascia, is essentially alike, though there may be some slight differences between them. The outwardly situated second fascia is simple in the present species while in *Wormaldi* it is furcate in its lower half, and in *striata* its upper half is wanting, the lower furcate portion alone running on the posterior margin; the 3rd fascia of the present species is furcate in its lower half, while that of *Wormaldi* is straight and simple and that of *striata* curved and simple though inclined to join with the above-stated marginal portion of the second fascia. Near the apex, there is a short fascia in the 3 species; it is nearest to the apex and therefore shortest in *Wormaldi*, *striata* comes next, and in the present species it is most removed from the apex and therefore longest. Besides there is in this species a series of short apical striae between each vein, which are never present in the other two species. Of course it is possible that the wing markings of the present species may sometimes break up into pieces which are often hardly possible to trace. Besides, the wing of *Wormaldi* is broader in proportion near apex while that of the present species is only moderately so and therefore it looks very slender.

6. ***Panorpa irregularis*** n. sp. (*Midare-shiriagemushi*).

(Pl. XI. figs. 7, 7a, 7b, ♂.)

Body deep black; rostrum and antennae black; palpi piceous; legs greyish or fuscous yellow with fuscous tarsi.

Wings broad, whitish, elliptical at the apex, a very broad black fascia beyond the middle, with the inner edge rather sharply defined; the fascia usually furcate externally just beyond the middle, forming a narrow branch ending on the posterior margin, oblique in a direction contrary to that of the fascia, so that there is in most cases a vitreous space enclosed between the fascia and the branch; in a few cases, however, the fascia has no branch and the part is sinuated or a certain portion of the branch remains on the posterior margin, forming an elongated marginal spot; two rather irregular black spots before the fascia, of which the hind one is usually

larger and of quadrate form; sometimes the two spots are united into an irregular narrow fascia; occasionally another series of two or three smaller spots before the last mentioned spots; these may also join with each other to form an irregular and incomplete fascia and in some case is connected with the fascia which is formed by the spots just-mentioned; apex also very broadly black, its inner margin sinuate; in some specimens, certain part of both the postmedial band and apical black space traversed longitudinally by a pale line between each successive veins; longitudinal veins mostly blackish and especially very conspicuous in the basal half; transverse veins mostly piceous in basal half and yellowish or testaceous in the space between the fascia and the apical portion.

Abdomen blackish; in the ♂ the posterior margin of the 3rd dorsal segment produced into a short median lobe just like that of *P. japonica*; 6th and 7th segments thick, cylindrical, truncate and equal in length; 8th slightly longer than the 7th, cylindrical; cheliferous segment short, the lateral pieces stout, the chelae brownish or piceous; appendages rather short, lineal, black, and slightly broader than in *P. japonica* with rather prominent ridges.

Expanse 32 mm.—36 mm.

A series of specimens captured at Goka-no-shō in Kiushiu by the author on May 27—29, 1908; a series of specimens presented to the author by the Imasu Primary School, obtained at Fuha-gōri, Gifu-ken, by children of the school, in Aug. 1908; two females captured on Mt. Hikosan, Buzen, Kiushu, by Baron TAKACHINO, on May 18, 1902.

This species is closely allied to the *P. japonica* of Thunberg, *P. pulchra*, *P. nipponensis*, *P. sinanoensis* of mine, and *P. dyscola* of NAVAS, recently described in 'Memorias d. l. Red Acad. d. Sienc. y. Artes d. Barcelona vol. VI, No. 25 (1908). From *P. nipponensis* and *P. sinanoensis* can be distinguished by the cheliferous appendages besides other points. To *P. japonica*, *P. pulchra* and *P. dyscola* it is very closely allied in the structural character though there may be found some slight differences in the form of the cheliferous appendages. The wing mark-

ing, upon which I chiefly relied for the distinction of species, show some differences between them, vid. in *P. japonica* there are in the male occasionally and in the female always found "two or three black spots before the (stigmatical) fascia"; in the present species, two spots are always present before the fascia and another series of two or three spots may occasionally be found before the spots or if wanting the spots just-mentioned may coalesce into a narrow fascia; in *P. pulchra* the two series of spots just mentioned are respectively united into a fascia and the two fasciae are joined posteriorly so as to show a V-shaped figure; besides there may occasionally be found some spots still internally, or this region may irregularly be suffused with black; in *P. dyscola*, though I have not yet seen the type specimen, the anterior fascia of the V-shaped figure found in *P. pulchra* is wanting, and the wing markings differ greatly in the sexes, while in *irregularis* and *pulchra*, there are no noticeable sexual differences. From these facts we may conclude that *irregularis* represents an intermediate form between *japonica* and *pulchra*, and closely allied to *dyscola* through the latter. The characteristic formes of the cheliferous appendages also appear to present broader and more acute than in *japonica* and those of *pulchra* are not so broad as in *irregularis* although the apex is acute as in the last species, and in both *irregularis* and *pulchra* the appendages appear slightly more divaricate than in *japonica*.

However these four species are very closely allied one with another and the differences above mentioned may be considered as differences of degree. I have therefore some grave doubts whether they may not all be amalgamated into one species. After much hesitation I have however finally made up my mind to recognize them as so many distinct species because differences of the same order as those found between them are used by recognized authorities in distinguishing other species of the same family, and moreover the above mentioned species occur respectively in detached places.

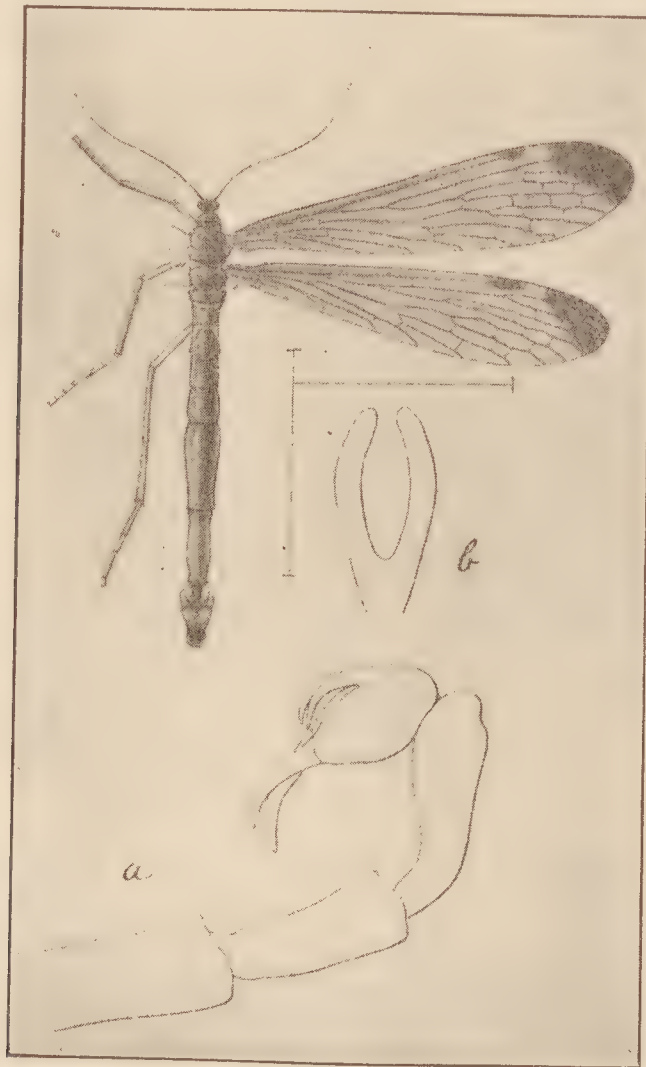
Anyhow the present species requires further study. If the above mentioned four species should prove to be one, I would not hesitate in proposing that *P. macrogaster* M'L. should also constitute a synonym of

it, because in my collection of *P. japonica* there is a series of specimens forming a transition to the former species, as, for instance, the partly fenestrated black markings and other characters.

7. *Panorpa chuzenjiensis* n. sp. (*Tsumaguro-shiriagemushi*).

Body black; rostrum black; antennae and palpi blackish piceous; legs fuscous ochraceous.

Wings moderate, tinged with ochraceous yellow; slightly narrower compared with that of *P. japonica*; apex elliptical; a small blackish costal



Panorpa chuzenjiensis n. sp. ♂. a. Apex of abdomen. b. Appendices.

patch somewhat triangular in the fore wing and semi-elliptic in the hind wing, just at the commencement of the pterostigma, occupying about one third of the latter; apex moderately tinged with blackish (not so broad as in *P. japonica* and its allies), with almost straight internal edge running obliquely posteriorly and outward; no other patch or fascia pres-

ent; pterostigma very slightly opaque; longitudinal veins black; transverse veins black except in the apical portion, where they are white.

Abdomen black; in the ♂ the posterior margin of the 3rd dorsal segment is produced into a short but very broad median lobe (broader than that of *P. japonica* and sometimes even that of *P. ochraceopennis mihi**); 6th, 7th and 8th segments cylindrical, almost equal in length and much slender than in *P. japonica* or its allies; 9th rather smaller and far less stout than in *japonica*; chelae long, the appendages rather long and moderately curved as in *P. Klugi* and *P. ochraceopennis* and the distal half bent downwards between the two lateral pieces.

Expanse 38 mm.

Two male and two female specimens obtained at Chuzenji, Nikkō, on July 19—22, '09.

This species is decidedly a distinct form in not having the broad submedian fascia or other series of striae which occur in all species of our *Panorpa*. Though in the structure of the cheliferous appendages it resembles to a great extent *P. Klugi* and *ochraceopennis*, a minute study always brings out the differences between them.

8. *Panorpa Wormaldi* M'Lach. (*Kiashi-shiriagemushi*).

(Pl. XI. figs. 8, 8a, 8b, ♂.)

Description of ♂.

Body black, except the ventral side of the thorax, the lateral sides of the abdomen and the apical part of the cheliferous segment, which are yellowish ochraceous in colour; rostrum and legs yellowish ochraceous; antennae black.

Wings marked just as in ♀., which was described by M'Lachlan in 'Trans. Ent. Soc. Lond., 1878, p. 186,' hind wing slightly shorter than the fore wing.

* I have stated in this paper that the lobe of *P. ochraceopennis* is broader than that of *P. japonica*. See p. 191. The lobe of *chuzenjiensis* is mostly equal to that of *ochraceopennis* but sometimes the latter is slightly broader.

Abdomen black; the lateral sides of the 1st to 5th segments yellowish ochraceous; 3rd abdominal segment produced in its posterior margin into a short broad median lobe; 2nd to 7th segments cylindrical, almost equal in length and width; 8th very slightly longer than the preceding segment, cylindrical, and slightly narrower; 9th segment yellowish ochraceous with the lateral pieces above tinted with piceous; lateral pieces stout, somewhat elongate; chelae very short, dilated towards its basal part; appendages rather larger, piceous black, broad, almost straight, somewhat dilated towards the apex which is abruptly acute.

Expanse 27 mm.

I have captured two males and females on Takaoyama near Hachiōji, in May of last year and in the same month of 1908. Prof. KINCAID and some friends of mine have also captured the species in the same month at the same locality. So that Takaoyama may be considered as a definite locality for this species.

9. *Panorpodes apicalis* n. sp. (*Tsumaguro-shiriagemushi-modoki*).

(Pl. XI. fig. 4, ♀.)

Body totally ochraceous; palpi and antennae testaceous; eyes ochraceous; the region of head including ocelli fuscous; legs yellow.

Wings pale ochraceous with the apex rounded; towards apex they are much broader in proportion than in *Panorpodes paradoxa*; apex suffused narrowly with fuscous; pterostigmatic region uniformly coloured as the rest of wings and not so opaque.

Expanse 33 mm.

A single female specimen in the collection of the Agricultural College captured by Prof. SASAKI on Takaoyama on May 23rd '03.

This species is also allied to *Panorpodes paradoxa* M'L., but differs by its lighter coloured and relatively broader wing with rounded apex, and uniformly coloured ptersotigma, *paradoxa* having deeply coloured wings with slightly narrowed elliptical apex and opaque testaceous pterostigma.

10. *Panorpodes singularis* n. sp. (*Kasuri-shiriagemushi-modoki*).

(Pl. XI. fig. 7, ♀.)

Body testaceous; rostrum with the palpi and antennae testaceous; legs yellowish.

Wings wholly pale yellowish with the apex rather acute; in the fore wing an irregular fuscous patch is present at the costal margin running in the anterior half of the wing; another small irregular patch near the posterior margin; a likewise fuscous patch more inwardly on cubital vein near the posterior margin; in both wings, a fuscous patch just at the costal margin covering the inner part of the pterostigma; the remaining (outer) part of the pterostigma yellowish; a light fuscous apical patch.

Abdomen slightly shorter than the ordinal *Panorpa*.

Expanse 32 mm.

A single female specimen in the collection of the Agricultural College, captured by the author on Hibaratōge, Yamagata-ken, July 24, '08.

This species has a singular form and no mistake is possible in its identification; still the species is to a certain degree allied to *P. decorata* M'L., in which a Y-shaped fuscous patch is present in both wings; if in the present species the patches of the fore wing should be more pronounced and connected with one another they should surely constitute a Y-shaped patch similar to that of *P. decorata*. It is however remarkable that those patches are actually absent in the hind wing except the pterostigmatal patch. If all the patches except the apical were absent a condition found in *P. apicalis* would result. Again if all the patches were wanting a form very near to *P. paradoxa* would result.

P.S. *Panorpa rectifasciata* mihi described in my former paper appears to be closely allied to *P. leucothryia* of NAVAS described from China in the work just referred to. They can be distinguished in many points,

best of all by the fact that in *rectifasciata* the pterstigmatal band is not so oblique and never bears spots between it and the apical dark portion.

I have captured a very large female specimen of *rectifasciata* at Nasu, Tochiki-ken, on July 19, '08. Expanse of wing 46 mm. It is the largest specimen of *Panorpa* I have seen so far.

Jan. 1910.

EXPLANATION OF PLATE XI.

(*Panorpa*; *Panorpodes*; *a* denotes apex of ♂; *b*, *c* appendices.)

- Fig. 1. *Panorpa ochraceopennis* n. sp., ♂. (1*a*, 1*b*).
 Fig. 2. *P. obscura* n. sp., ♂. (2*a*, 2*b*). (2*c* appendices of *P. rectifasciata* Miyake).
 Fig. 3. *P. gokaensis* n. sp., ♂. (3*a*, 3*b*).
 Fig. 4. *Panorpodes apicalis* n. sp., ♀.
 Fig. 5. *Panorpa multifasciaria* n. sp., ♀. (5*a*, 5*b* chelae).
 Fig. 6. *P. magnicauda* n. sp., ♂. (6*a*, 6*b*).
 Fig. 7. *P. irregularis* n. sp., ♂. (7*a*, 7*b*).
 Fig. 8. *P. Wormaldi* M'Lach., ♂. (8*a*, 8*b*).
 Fig. 9. *Panorpodes singularis* n. sp., ♀.
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Some Notes on the Arctianae of Japan.

BY

T. Miyake.

With one Figure in the Text.

On the Arctianae of Japan, I have hitherto published two papers, one¹ in 1906, describing 27 species, and the other² in 1909, enumerating 32 species. Since then I have been favoured by many of my correspondents with various consignments of the moths, including three species, two of which I consider to be new to Japan and the other new to science. They are therefore described in the following lines and I may utilize this opportunity by adding remarks on some larvae which have not yet been mentioned in any scientific publications. Two of them feed on the mulberry-tree, so that we have in all nine species of the present subfamily injurious to the mulberry.

Pericallia matronula L. (*Jōzan-hitori*).

Noctua matronula L., "Syst. Nat., i, p. 509 (1758);"* id, ii, p. 835 (1767).

Phalaena idriensis Scop., "Annus N.H., v, p. 113 (1772)."

Bombyx matrona Hübn., "Eur. Schmett., ii., figs. 238, 239 (1800)."

Pericallia matronula Hampson, Cat. Lep. Phal., iii, p. 360 (1901);
Stand., Cat. Lep. pal., p. 371 (1901).

1. MIYAKE: Tōga-akwa ni kwansuru Kenkyū hōkoku, Extra reports from Imp. Cent. Agr. Exp. Station, No. 22 (1906).

2. MIYAKE: A revision of the Arctianae of Japan, Bull. Coll. Agr. Tokyo Imp. Univ., vol. VIII, No. 2 (1909).

* The literature which are not accessible to me are indicated with " ".

Many years ago I received a fair male specimen of this species from my friend Mr. Oguma, who obtained it from one of his friends, the locality being said to be somewhere in the northern part of the Main Island. As nothing certain was known about the capturer, or the date of capture, or the locality, I have long hesitated to count the specimen as belonging to our fauna. Last summer however I found in the Sapporo Agricultural College a series of this species, with exact record of the locality and the date of capture; and I was also favoured with a specimen through the kindness of Prof. MATSUMURA of the College. They were all captured at Jōzankei, a mountainous place with a hot spring, about 16 miles from Sapporo. The specimens were mostly collected in the month of August. On examining the specimens and comparing them with the typical European form, there is no doubt that the present specimens are referable to *Pericallia matronula*, though I am not acquainted with any series of European specimens connecting them with the Japanese form. Our specimens showed some variations which are likely to occur also among European forms, after all however fore wings seem to me to be much darker in the Japanese than in the European form.

2. ***Diacrisia obliquizonata*** n. sp. (*Futo-sujimon-hitori*).

♂. Very pale buff; palpi black at extremity, crimson at base; frons fuscous; antennae biserrate, black; femora crimson above, tibiae and tarsi brownish; pectus suffused with fuscous on its middle; abdomen crimson dorsally except at base and extremity, where it is somewhat lightly coloured, with dorsal, lateral and sublateral series of black spots.

Fore wing with a black fascia on base of costa expanding into an antemedial spot; a black spot in the discoidal cell just below the above-mentioned spot; another antemedial spot above vein 1; just below the last-mentioned spot a broad black bar is present, occupying entirely the interspace between vein 1 and the inner margin and reaching to the middle of the latter. An oblique broad black fascia, interrupted by the veins that traverse it, from apex to the middle of the inner margin, uniting with the

above-stated bar. A short black streak on the costal margin just beyond the middle; another small triangular spot just behind the streak; an obscure black discoidal spot.

Hind wing whitish, with the inner margin slightly tinged with buff; a round black discoidal spot; a black spot at the anal angle; a somewhat elongate subterminal black spot on the posterior side of vein 2; another much smaller spot on the anterior side of the same; two spots, of which the posterior one is smaller, respectively on each side of vein 5; spots of the hind wing somewhat paler in coloration.



Diacrisia obliquizonata n. sp.:

- a. Male (natural size). b. Antenna of male (magnified).
c. Venation of wing (magnified).

Underside of wings much paler in coloration with similar but more lightly coloured markings; the discoidal spot of fore wing slightly more distinct; some crimson hairs on base of the fore wing. Expanse 42 mm.

♀. Much larger in size with almost similar coloration and markings; wing markings, especially of fore wing, much more pronounced than in the male specimen; a black antemedial spot in discoidal cell and a small triangular spot below the postmedial costal streak, which are found in the male specimen, entirely obsolete. Expanse 52 mm.

A male and a female specimen were kindly given me by Mr. Niwa of the Tokyo Sericultural Institute. They were sent to him in the state

of larvae as feeding on mulberry leaves, by Mr. N. Furuya of Hatsu-shika, Tajima, Hyogo Prefecture, in September 1908. Mr. Niwa told me that the larvae pupated soon after reaching him, so that he could not make observations on them. The moths emerged in the month of May of this year.

This year Mr. Niwa wrote to the same person for the same larvae and received certain caterpillars in large number. They undoubtedly belong to a certain Arctid moth, which I have not yet seen nor can identify with any of the published descriptions. However I cannot but hesitate to regard the caterpillars as those of the present species, because they were too numerous in the locality to be referred to such a rare moth as the present, described here for the first time. Mr. Niwa told me that these larvae looked very like those he saw last year, that he can not discover any difference between them, though he is not sure of their specific identity. Of our commonest Arctid moths, the species whose larva is still unknown, is *Diacrisia seriatopunctata*, to which the present species is much more closely allied than the remaining species. I have therefore an idea that the caterpillars in question, if they do not belong to the present species, may possibly be those of *seriatopunctata*. Anyhow the caterpillars are being reared by Mr. Niwa and the issue will come out next year.

Description of the larva:—

Early stages. Head black, body fulvous; tubercles black; hairs rather short, white mixed with black; legs piceous. Before moult it becomes dusky.

Later stage. Head black; body blackish; thoracic segments with black hairs; first three abdominal segments with fulvous hairs; the remaining abdominal segments laterally with fulvous, and dorsally and ventrally with black hairs; thoracic and last abdominal segments bear some long whitish hairs; tubercles purplish black; legs piceous.

Food-plant: mulberry-tree.

Though the present species is, as already mentioned, allied to some extent to *seriatopunctata* still we can not consider the one as the aberrant

form of other. They are different in the following points: the antennae of male of the present species is biserrate while in *seriatopunctata* they are bipectinate. The colour of thorax and wing in this species is very pale buff, while in *seriatopunctata* it is very deeply buff, and moreover in male usually tinged with crimson or brown. A series of small black spots are found in the fore wing of *seriatopunctata* running from apex to the inner margin beyond the middle, while in the present species the serial black spots become a very broad conspicuous band, which is only interrupted by the veins which pass it, and terminates at the middle of the inner margin. Besides there is a bar at the middle of hind margin in *obliquizonata* which is absent in *seriatopunctata*.

3. *Diacrisia inaequalis* Butl. (*Kakumon-hitori*).

Spilarctia inaequalis Butl., Ann. Mag. Nat. Hist., (5) IV, p. 351 (1879).

Spilosoma inaequalis Leech, Proc. Zool. Soc. Lond., 1888, p. 619.

Thyrgorina inaequalis Leech, Trans. Ent. Soc. Lond., 1899, p. 159.

Diacrisia inaequalis Hampson, Cat. Lep. Phal., III, p. 288, pl. XLV, fig. 9 (1901); Miyake, Bull. Coll. Agr. Tokyo Imp. Univ. Vol. VIII, No. 2 (1909).

This species has been described by many authors from Japan, as shown by the above references. The larva and the life-history however, I believe to be given here for the first time. The larva was discovered by Mr. Y. Mukōgawa of Namisemura, Isshi-gun, Miye Prefecture, as an insect injurious to the mulberry-tree in that locality. He has kindly sent me some of the larvae and I am under deep obligation to him to be able to furnish the following description.

Description of the larva:—Head reddish ochreous; body dorsally dark brown with slight purplish tinge, laterally ochreous mottled with brown patches; cervical shield shining metallic blue-green; dorsal and subdorsal tubercles metallic blue; lateral, subspiracular and basal tubercles ochreous; each tubercles with hairs of black and white; tubercles of 4-9 segments mixed besides with brownish hairs; legs testaceous, abdominal legs with

a longitudinal fuscous streak, the basal part of which tinged with metallic blue; spiracles grey with black ring. Food-plants: mulberry-tree.

Pupa:—Blackish brown.

According to Mr. Mukogawa's statement, the insect has two generations in a year; it passes the winter in the state of larvae of the third or fourth stage, and moulting three or four times in the next spring; pupation takes place at the beginning of May, and at the end of the month or beginning of June the adult emerges and deposits eggs, which hatch out in about two weeks and pupate at the middle of July; at the end of August or at the beginning of September the second adult brood emerges. The injury to the mulberry is said to be rather serious.

4. *Apantesis proxima* Guer. (*Amerka-hitori*).

Chelonia proxima Guer., "Icon. R. Anim., III, p. 514 (1844)."

Apantesis proxima Hampson, Cat. Lep. Phal., III, p. 44 (1901).

I have received a male specimen from my friend Mr. Takano of Yokohama, an enthusiastic lepidopterologist, who captured it himself in the neighbourhood of his house. The moth well agrees with Hampson's and many other American authors' descriptions of the above species and there is no room for doubt in referring it to the present species. However as I am not aware of the occurrence of this species anywhere in Japan or in the adjacent localities, and as it does not seem to have any palaearctic affinity so far, its singular occurrence in this case may rather be due to some accidental transportation, as Yokohama harbours many ships—many of them from the United States.

Nov. 1909.

The Mantispidae of Japan.

BY

T. Miyake.

With Plate XII.

Of that small family Mantispidae consisting of a few genera only one species, *Mantispa japonica*, has hitherto been described from Japan by M'LACHLAN. Some other species of this very rare and small family have however been discovered lately from various parts of Japan, but they have, so far as I know, remained undetermined.

Through the kindness of Mr. NAWA, Chief of the Nawa Entomological Laboratory of Gifu, his valuable collection of Japanese Mantispidae, containing four species, was placed at my disposal. This, together with the collection of the Agricultural College of Tokyo and of mine, has enabled me to get a general view of this family in Japan.

All these species, five in number, seem to belong to the genus *Mantispa*, and three of them are new to science and one new to Japan. They are:—

Mantispa magna n. sp.

M. Nawae n. sp.

M. Sasakii n. sp.

M. japonica M'Lach.

M. 4-tuberculata West.

Prof. MATSUMURA of the Agricultural College of Sapporo has described a new species *Mantispa diminuta* in his "Konchū Bunruigaku" (Systematic Entomology) vol. 1, p. 169 (1897), but his description is

too brief to allow a satisfactory identification, though there is a specimen which apparently agrees with it.

Besides the descriptions of the three new species I have reproduced in this paper the original descriptions of the other two species, as well as that of Prof. MATSUMURA's new species, in order to bring out some doubtful points in the original descriptions and to facilitate the work of future students.

For systematic purposes the male genital parts have mostly been used in Neuroptera, these are available only where male specimens are present and moreover they must be new and perfect. In the present case, some species are represented only by females and where males are present, the genital parts are shrunk, dusty or imperfect, so that I had to depend on other characters.

Lastly the author takes the pleasure of expressing his hearty thanks to Mr. NAWA, who has kindly lent his precious specimens for examination. Thanks are also due to Mr. YANO of the Science College and to Mr. KINOSHITA of the Second High School who have favoured me with some specimens. To Prof. SASAKI and Prof. GOTO I am also under deep obligation for the kind advices rendered me during the study.

***Mantispa magna*. n. sp. (*Ō-kamakiri-modoki*).**

(Pl. XII. figs. 3, 3a, 3b, 3c, ♀.)

Head ochraceous yellow, nearly flat on the crown, slightly impressed on each side from the base of the antennæ along the margin of the eyes; clypeus and a transverse line between the base of the antennæ black; a transverse brown bar across the top of the head; labrum elliptic; maxillary and labial palpi fulvous; antennæ about two times the length of the head, fuscous with the basal two and terminal three joints ochraceous, moderate sized, 40-jointed; second joint scarcely larger than the third, the remaining joints except the first to third very short and transverse. Prothorax long, dark fulvous, transversely rugose except the anterior dilated portion; another dilated portion beyond the middle; two ordinal tubercles

behind the anterior portion; two longitudinal elongated x-formed fuscous black lines edged outwardly and posteriorly with obscure fulvous line commencing vaguely at the anterior dilated portion and ending at the angles of the posterior dilated portion; a faint fulvous median line rather conspicuous between the two tubercles; scutellum fuscous black; underside dark fulvous with an indirect narrow median fulvous line. Meso- and metanota reddish fulvous, with scutella ochraceous; lateral and underside reddish fulvous. Abdomen reddish fulvous above, with the first segment* very short; second segment* *produced at middle on the hind margin; a broad ochraceous median streak from beyond the hind margin of the first segment to the angle of the produced portion of the second segment; third segment posteriorly margined with a low triangular ochraceous patch which is anteriorly lined with λ -shaped indistinct fuscous black streak; fourth segment with much smaller but higher triangular ochraceous patch just in opposition to the triangle of the preceding segment surrounded laterally with fuscous patches; the apex of the triangular patch produced somewhat posteriorly along median line; hind margins of the third and fourth segments tinged broadly with fuscous; the remaining terminal segments rather ochraceous in colour with longitudinal fuscous band on each side; underside fuscous. Fore legs uniformly dark fulvous; coxæ with a whitish constricted ring about one third from the base; femora with the extremity and spiny margin varied with orange colour; the longest basal spine reddish fulvous with the apex fuscous; tibiae with an obscure fuscous streak about the middle one third of the upper edge; lower edge broadly fuscous; tarsi with the basal joint elongate-conic, fuscous and hairy beneath; of the four terminal joints the last one is longest and terminated by a single unguis; mid- and hind-legs moderately long, fulvous; femora and tibia of the mid-leg with the whole of the former and the base and a line along upper side of the latter dusky; hind leg with the extremity of the femora and with the

* ** I think this way of counting the number of abdominal segments is natural, though the first segment of this numbering is very short, and the second segment seems to form a single segment with the third.

basal one third of tibiae dusky; unguis of both legs terminated by six acute teeth. Wings long, nearly colourless and hyaline with many minute elongated fulvous veins so as to form a great number of discoidal cells; pterostigma of both wings ochraceous fuscous; the base of fore wing and anterior margin of both wings from base to apex suffused with light fulvous brown; sixteen oblique mostly elongated discoidal cells dependent on the ordinary radial sectors, the veinlets dividing the cells almost straight.

Expanse of wings 64 mm; length of body 27 mm.

Two female specimens: one in the collection of the NAWA Entomological Laboratory, captured by Mr. NAGANO in Fukuokaken, Kiushiu, April '02; one in the collection of the Agricultural College, captured by Baron TAKACHIHIO, on rice-plant at Soeda, Fukuokaken, Kiushiu, Oct. 12, '05. This is undoubtedly the largest species of the genus in Japan and presumably one of the largest of the whole family. It is allied to a certain extent to *Mantispa arcolaris* of WESTWOOD, but is readily to be distinguished in many respects.

***Mantispa Nawae* n. sp. (*Kamakiri-modoki*).**

(Pl. XII. figs. 4, 4a, 4b, ♀.)

Head entirely chestnut with rather deep impression between the antennae and the margin of the eyes; a well-defined central carina with a fuscous tubercle on each side; clypeus dusted with fuscous; labrum triangular, maxillary and labial palpi ochraceous yellow; antennae fuscous, longer than twice the length of the head; basal joints fulvous; 38-jointed. Prothorax greyish-ochraceous transversely sulcated; the second and third grooves from the border of the thorax connected along the median line by a longitudinal groove so as to form I-shaped impression; the dilated anterior portion diffused with fuscous; tubercles rather undeveloped; meso- and metathorax ochraceous yellow with two brownish patches on the anterior margin of the former. Abdomen fuscous above with an obscure central fuscous band, piceous beneath. Legs ochraceous

with the anterior raptorial femora and tibiae dusted with fuvous brown. Wings colourless, hyaline with blackish fuscous veins; costa and radius ochraceous tinted with fuscous in some part; pterostigma long and narrow, blood-red; veins near apex of the wing also tinted with blood-red; thirteen oblique elongated discoidal cells dependent on the ordinary radial sectors, the veinlets dividing them slightly bended.

Expanse of wings 54 mm; length of body 27 mm.

A single female specimen in the collection of the NAWA Entomological Laboratory from Mt. Ibukiyama, Hida, Aug., 1892. I take great pleasure in naming this species after Mr. NAWA, Chief of the NAWA Entomological Laboratory at Gifu, by whose kindness I was enabled to examine this valuable specimen.

Mantispa Sasakii n. sp. (*Ki-kamakiri-modoki*).

(Pl. XII. figs. 2, 2a, 2b, ♀.)

Head ochraceous yellow, with impression on each side of the insertion of antennae uniting transversely on the forehead and giving rise to an elevated central carina behind it; the transverse portion connecting the base of antennae varied with testaceous; the impression between the base of antennae and the margin of eyes fuscous; antennae fuscous, long and slender, with 31 joints.* Prothorax ochraceous yellow, transversely rugose with no markings; the dilated anterior portion fulvous brown; the two tubercles rather inconspicuous; meso- and metathorax ochraceous yellow, the former varied with slight fuscous above; abdomen fulvous varied with fuscous; basal segment piceous. Legs ochraceous with the anterior raptorial femora and tibiae tinted with fulvous. Wings colourless and hyaline with fuscous brown veins; costa and radius ochraceous; pterostigma narrow and long, blood-red; twelve rather elongated discoidal cells dependent on the third sector of the radius in the fore wings, and thirteen on the second sector in the hind wings; most of cells bended and some narrowed in the middle through curving of the veins.

* I have actually counted 31 joints as here stated, but I have some suspicion that the terminal joint has been dropped; in that case it must have possessed originally 23 joints.

Two female specimens: one captured by the author on Mt. Daimanji in Oki-island on Aug. 14, '06, one in the collection of the Agricultural College, captured at Urami-no-taki road in Nikko on Aug. 28, '95.

I have named this species in honour of Professor SASAKI of the Agricultural College, Tokyo Imperial University.

Mantispa 4-tuberculata West. (*Tsumaguro-kamakiri-modoki*).

(Pl. XII. figs. 1, 1a, 1b, ♂.)

Trans. Ent. Soc. Lond., new series vol. 1, p. 264, pl. 18, fig. 1 (1852).

Original descriptions as follows:— "Bruneo-fulva, flavo nigroque varia, antennis brevissimis, 32-articulatis, pronoto carina transversa ante alteraque pone medium alarum, stigmat venisque subcostalibus fulvis, nubilaque apicali fusca."

"Long. corp. lin. 5—9; expans. alar. antic. 11—17."

"Habitat Northern India. Mus. W. W. Saunders."

"This is a very elegant species, nearly allied to *M. auriventris* of Gerérin Méneville. The head is bright yellow, nearly flat on the crown, slightly impressed on each side at the base of the antennæ; the clypeus and a transverse line beneath the base of antennæ black, and at transvers *brown* bar across the top of the head; labrum nearly circular; palpi fulvous; antennæ scarcely more than one and a half times the length of the head, thick, fulvous, basal joint yellow, moderate sized; second joint small, scarcely larger than the third, remaining joints, especially beyond the middle, very short and transverse. Prothorax dark fulvous, deeply transversely sulcated, forming a more strongly marked carina before and another behind the middle; anterior part semicircularly dilated in front, yellow, with the anterior margin black, and a *bruneous* transverse fascia at its hinder part; meso- and metathorax dark fulvous, with the scutella yellow. Abdomen above with the basal half dark fulvous, the remaining half bright yellow; the second and third joints with a broad black hind margin; sides of the abdomen yellow; the three terminal segments blackish; beneath dark fulvous; fore legs dark fulvous; femur

yellow on the outside, dark brown on the inside, large spine yellow, placed nearly in the middle of the thigh; four hind legs fulvous; tibiæ paler, with a dusky broad ring near the base; ungues short, broad, terminated by four or five sharp teeth; pulvillus broad; wings narrow; principal veins fulvous; stigma long, orange brown; fore wings suffused at the base, and all the wings with an apical cloud of fulvous brown; all the wings with from *nine to eleven oblique discoidal cells* dependent on the ordinary radial sectors, the veinlets dividing the cells slightly curved."

Two specimens are in my hand; one male specimen captured by Mr. KINOSHITA at Koshigoye near Kamakura, Sept. 23, '07 and the other (sex undeterminable) specimen from the collection of Mr. NAWA captured at Inaba-gun, Gifu-ken, Aug., 1892. Both specimens agree very closely with the above description of *Mantispa 4-tuberculata* of WESTWOOD, except in the points (distinguished by italics), i. e. the transverse bar across the top of the head is *black* instead of *brown*; the transverse fascia at the hinder part of the anterior dilated portion of prothorax is *black* instead of *brunneous*; the cells dependent on the ordinary radial sectors are *eleven to thirteen* instead of *nine to eleven*. Besides, the apices of the wings are more strongly suffused than in *4-tuberculata*, as given by WESTWOOD in his description and figure. The abdomen is also slightly different in colour though only in the degree. All the veins have minute hairs on them which fact is not mentioned in the description of WESTWOOD. On the ground of these differences, I had at first considered the present specimens to belong to another and a new species. After further reflection however I have thought it more natural to refer them to *4-tuberculata* and to regard the above differences as local variations or at most as varietal characters. Anyhow it is a very interesting and remarkable fact that the same or very closely allied species occurs in Northern India and the interior of the Main Island of Japan, localities so far apart from each other.

Mantispa japonica M'Lach. (*Hime-kamakiri-modoki*).

(Pl. XII. figs. 5, 5a, 5b, ♂.)

Trans. Ent. Soc. Lond., p. 1875, p. 178.

Description: "Head yellow, with a black line down the face; antennæ fuscous, the basal joints fuscous; sub-moniliform, with about 30 joints; palpi reddish, the terminal joint piceous at the apex. Prothorax long, dark brown, the dilated anterior portion black, with two yellow spots, forming a nearly continuous transverse band; a little behind the anterior portion are two yellow tubercles, the remaining portion finely corrugate. Meso- and metathorax varied with yellow, black, and brown. Abdomen much thickened at the apex, yellow above, with an irregular central brown band; beneath blackish, varied with yellow; in the ♂ there are two short, stout, and obtuse lateral appendages, and a large boat-shaped lobe from the middle of the last ventral segment, from within which proceeds the spiniform penis, which is strongly curved, and annulated with black and testaceous. Legs yellowish; posterior tibiæ marked with brownish externally, and the tarsi are brownish; anterior raptorial femora deep black internally, strongly toothed, the basal spine very long. Wings vitreous; neuration black; costa and radius pale; pterostigma very long and narrow, blood red; 7—8 costal nervules in the anterior wings, and about 12 discal cellules, the greater part of which are narrowed in the middle through the bending of the nervules."

"Expanse 29 mm."

"One ♂ from Yokohama (Pryer), in WORMALD's collection."

I have a male specimen from Mr. YANO, captured by him on Mt. Kirishima, Kiushu, July 31, '08, which agrees well with the description. There is also a male specimen in the collection of Mr. NAWA. Both specimens however are smaller than the typical form, the expanse of wings of the former being 26 mm. and that of the latter 24 mm.

Mantispa diminuta Mats. (*Chibi-kamakiri-modoki*).

MATSUMURA, "Konchiu-bunruigaku" (Systematic Entomology), vol. 1, p. 169 (1907).

"Allied to *Mantispa japonica*, but smaller, antennæ blackish fuscous, the basal part yellow; T-shaped blackish fuscous mark on the vertex, occiput dark fuscous; legs yellowish white; length of body 8 mm.; expanse 22 mm."

There is a specimen (sex undeterminable) in the collection of the Agricultural College, captured at Karuisawajiri, Iwashiro, Aug., '02, which agrees apparently with the above description, but the two simple description does not enable me satisfactorily to determine it, the more so as I can not recognize the specific difference between *japonica* and *diminuta*. Until a more precise description of *diminuta* is published, I shall have to look upon the specimen provisionally as merely an aberrant form of *japonica*.

Feb., 1909.

EXPLANATION OF PLATE XII.

(*Mantispa*; *a* denotes basal and terminal joints of antennae; *b* extremity of leg).

- Fig. 1. *Mantispa 4-tuberculata* West. ♂. (1*a*, 1*b*).
 Fig. 2. *M. Sasakii* n. sp. ♀. (2*a*, 2*b*).
 Fig. 3. *M. magna* n. sp. ♀. (3*a*, 3*b*). (3*c* I, II and III abdominal segments).
 Fig. 4. *M. Nawae* n. sp. ♀. (4*a*, 4*b*).
 Fig. 5. *M. japonica* M'Lach. ♂. (5*a*, 5*b*).
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Some Experiments on Bombyx Mori.

BY

R. Inouye.

With two Figures in the Text.

A. On the Quantities of the Mulberry Leaves given to Silkworms and their Litters.

About twenty years ago, some investigations on this subject were carried out by Dr. O. KELLNER¹ upon the silkworms of the spring race. Last summer (1908), I made some experiments on the same subject upon a summer race Awojiku (bivoltine).

On the 12th July, the young worms of Awojiku came out for the first time. I separated them into two lots and marked them respectively A and B, and reared them on separate trays. In A, I counted 589 worms, and in B, 488. The weight of A was 0.236 g. and that of B 0.171 g.; the average weight of ten young worms was 0.0038g.

In the first stage the worms were fed with finely chopped young leaves with the veins entirely removed. They grew vigorously, and after five days, all of them began to undergo the first moulting.

The average temperatures and humidities in this stage were as follows:

Worms of 1st stage.	Indoor.		Outdoor.	
	Average Temperature.	Average Humidity.	Average Temperature.	Average Humidity.
1st day	78°.6 F	70.7	77° F	72.4
2nd day	76° „	78	74° „	81
3rd day	77° „	75.5	74°.6 „	79
4th day	75° „	79	72° „	83
5th day	76° „	84	74° „	92

1. Landw. Versuchsstat. Bd. XXX, 1882, p. 59 u. Bd. XXXIII, 1886, p. 35.

In the second stage, the worms were also fed with chopped young leaves with their hard veins removed. The worms grew well and began to undergo the second moult, four days after the completion of the first moulting (the 20th July.).

The average temperatures and humidities during this stage were as follows:

Worms of 2nd stage.	Indoor.		Outdoor.	
	Average Temperature.	Average Humidity.	Average Temperature.	Average Humidity.
1st day	80° F	76	79° F	73
2nd day	75° „	78	74° „	76
3rd day	73° „	80.2	70° 7 „	82.1
4th day	74° 6 „	78.2	74° 4 „	77.4

In the third stage, large soft leaves with the petioles removed, were given in pieces a little larger than in the previous stage. In this stage, the diseased worms were scrupulously picked out. The third moulting began on the 24th July.

Worms of 3rd stage.	Indoor.		Outdoor.	
	Average Temp.	Average Humidity.	Average Temp.	Average Humidity.
1st day	78° 5 F	82.3	75° F	87
2nd day	78° „	79	78° „	82
3rd day	75° „	84	72° 8 „	86
4th day	77° 6 „	82.7	76° 3 „	84.1

On the 15th July, the third moulting was completely finished. At the beginning of the fourth stage, the total number of worms was reduced to 500. On the 30th July the last moulting began.

Worms of 4th stage.	Indoor.		Outdoor.	
	Average Temperature.	Average Humidity.	Average Temperature.	Average Humidity.
1st day	83° 6 F	73.3	79° F	82
2nd day	84° „	76.1	82° „	67.7
3rd day	83° 3 „	78.5	80.6 „	78.8
4th day	83° „	74.8	81° 6 „	76.7
5th day	84° 3 „	71	84° 3 „	70

During the 5th age, entire leaves were given to the worms. After two days of this stage, many of them died from flacherie, but the lost individuals were supplied always with healthy ones. On the 5th day, the weight of the worms reached the maximum; the average weight of ten individuals being 27.05 grams (7.2 momme). On the 6th August, the worms attained maturity and commenced to spin cocoons. The average weight of ten mature individuals came down to 25.55 grams (6.8 momme).

Average temperature and humidity during the fifth stage.

Worms of 5th stage.	Indoor.		Outdoor.	
	Average Temperature.	Average Humidity.	Average Temperature.	Average Humidity.
1st day	83°.6 F	78.6	82°.8 F	77.6
2nd „	83°.4 „	77.8	82°.2 „	77.8
3rd „	83°.5 „	77	82°.5 „	76
4th „	80° „	74	78° „	75.5
5th „	80°.5 „	74.8	79°.2 „	74.5
6th „	81°.8 „	76.3	81°.8 „	79.3
7th „	80°.5 „	80	77°.5 „	89.5

The litters were collected twice during each stage (at the middle and end of each stage) and weighed; and after complete dessication they were again weighed. After the 3rd stage, the excrements were separated from the waste leaves and weighed. In each stage, the fresh leaves to be given were weighed in a dry state, as also the waste leaves left behind.

The water and dry matter of the mulberry leaves obtained were as follows:

Stage of silkworms.	Water.	Dry matter.
I	73.57 %	26.43 %
II	69.90 „	30.10 „
III	66.22 „	33.78 „
IV	65.48 „	34.52 „
V	68.30 „	31.70 „

The following shows the weight of the fresh mulberry leaves given to worms, and of litters per 1000 individuals.

Age.	The fresh weight of the mulberry leaves given to worms.	The dry weight of the same.	The air dry weight of litters.	The air dry weight of excrements.	
I.	g. 28.9	g. 7.6	g. 12.1	—	
	50 0	13.2	17.0	—	
	g. momme 78.9(21.0)	g. momme 20.8(5.5)	g. momme 29.1(7.7)	—	
II.	105.4	31.7	51.5	—	
	115.5	34.8	38.0	—	
	Sum 220.9(58.8)	66.5(17.7)	89.5(23 8)	—	
III.	278.4	94.0	71.3	22.8	
	369.7	124.9	103.0	30.7	
	Sum 648.1(172.4)	218.9(58 2)	174.3(46.4)	53.5(14.2)	
IV.	1126.4	388.8	287.6	168.2	
	1113.0	384.2	348.5	182.4	
	Sum 2239.4(595.7)	773.0(205.6)	636.1(169.2)	350.6(93.2)	
V.	1st and 2nd day	1773.3	562.2	418.8	176.0
	3rd day	1472.7	466.8	368.8	213.6
	4th day	1502.8	476.4	333.0	219.8
	5th day	2254.2	714.6	697.4	423.0
	6th day	1653.1	524.0	481.2	318.0
	7th day	736.4	233.5	361.1	182.4
	Sum 9392.5(2498.4)	2977.5(792.0)	2660.3(707.6)	1532.8(407.7)	
Total.	12579.8(3346.2)	4056.7(1079.0)	3589.3(954.8)	1936.9(515.2)	

The air dry weight of waste leaves.	The dry weight of litters.	The dry weight of excrement.	The dry weight of waste leaves.	The diff. between the dry matter of the given leaves and that of litters.
—	g. 7.3	—	—	g. 0.3
—	12.1	—	—	1.1
—	g. momme 19.4(5.2)	—	—	g. momme 1.4(0.4)
—	27.0	—	—	4.7
—	27.0	—	—	7.8
—	54.0(14.4)	—	—	12.5(3.3)
48.4	53.9	19.5	34.4	40.1
72.3	74.7	25.6	49.1	50.2
120.7(32.1)	128.6(34.2)	45.1(12.0)	83.5(22.2)	90.3(24.0)
119.4	240.0	139.9	100.1	148.7
166.1	278.1	147.5	130.6	106.0
285.5(75.9)	518.1(137.8)	287.4(76.4)	230.7(61.4)	254.7(67.8)
242.8	306.1	130.1	176.0	256.0
155.2	279.5	157.9	121.6	187.3
113.2	253.7	162.5	91.2	222.7
274.4	523.8	312.7	211.1	190.8
163.2	381.6	250.1	131.5	142.5
178.7	267.5	134.8	132.7	-34.0*
1127.5(299.9)	2012.2(535.2)	1148.1(305.4)	864.1(229.9)	965.3(256.8)
1533.7(408.0)	2732.3(726.8)	1480.6(393.8)	1178.3(313.4)	1324.2(352.2)

* It strikes me as very strange that in the above table the difference between the dry matter of the supplied mulberry-leaves and that of the litters, that is the digested part, on the last day of the 5th age is negative, but since, as the following table shows, the weight of the silkworms decreases on that day, it may be possible.

Daily weighing of ten 5th-age-worms.

1st & 2nd day.	3rd day.	4th day.	5th day.	6th day.	7th day.
g. mom. 7.514 (2)	g. mom. 11.42(3.04)	g. mom. 20.776(5.53)	g. mom. 25.93(6.9)	g. mom. 27.05(7.2)	g. mom. 25.55(6.8)

Conclusion.

The preceding table shows that 12579.6 grams (about 3350 momme) of fresh mulberry leaves, whose dry matter was 4056.59 grams, were needed for rearing 1000 silkworms, and 1324 grams (nearly 33 per cent.) of the given mulberry leaves were assimilated. The results are different from those of the similar experiments on the spring race carried out by Dr. O. KELLNER. The mulberry leaves which he gave to 1000 silkworms through their larval stages, weighed 5125.6 grams in a dry state, of which 2172 grams (nearly 42 per cent.) were digested by the worms. He took the precaution not to give too much leaves during the rearing, and the same precaution was adopted in my experiments. Hence, it may be said that the silkworm of the summer races, at least Awojiku, needs less mulberry leaves while growing than the worm of the spring races; and also it may be due to this fact that the summer races generally produce less silk than the spring races.

Next, the percentage weights of the supplied mulberry leaves and of the litters in each age were calculated with the following results:

(A)

Age.	The fresh weight of the supplied mulberry leaves.	The dry matter of the supplied mulberry leaves.	The air dry matter of litters.	The air dry matter of excrements.
I	0.63	0.51	0.80	—
II	1.75	1.64	2.49	—
III	5.15	5.40	4.86	2.76
IV	17.80	19.05	17.72	18.10
V	74.67	73.40	74.13	79.14
Total	100	100	100	100

(B)

Age.	The air dry matter of waste leaves.	The dry matter of litters.	The dry matter of excrements.	The dry matter of waste leaves.	Diff. between the dry matter of the suppl. mulberry leaves and that of litters.
I	—	0.71	—	—	0.10
II	—	1.98	—	—	0.94
III	7.87	4.71	3.05	7.09	6.82
IV	18.61	18.96	19.41	19.59	19.24
V	73.52	73.64	77.54	73.32	72.90
Total	100	100	100	100	100

From the above table it is seen that the consumption of the leaves during the fifth age amounted to more than 70 per cent. of the total of the five stages. Although the fifth age lasts for only one fourth of the total larval stages, the silkworm consumes more than twice the quantity of the food taken during the other ages.

In fact, since the growth of the silkworm takes place most vigorously in this stage, the hygienic precautions and climatal conditions during it may have a great influence on the silk crop.

B. On the Influence of Carbon Dioxide Gas upon Silkworm.

This experiment was carried out with the following apparatus.

I took a large belljar (Fig. 1. A), open at the top and having of 9360 c.c. The rim of its larger (lower) opening is greased usually with vaseline and put on a thick glass plate. *M* is a glass-tube twice bent on itself and filled with water, and serving as a manometer in order to indicate the pressure of the inside of the bottle A, when the latter is sealed up after filling with CO₂ gas; B is a caustic potash bulb, which is used to catch CO₂ gas; C is an apparatus for purifying the air by absorbing its CO₂. It is composed of several bottles *a*, *b*, *c*, *d* and *e*; *a* contains a solution

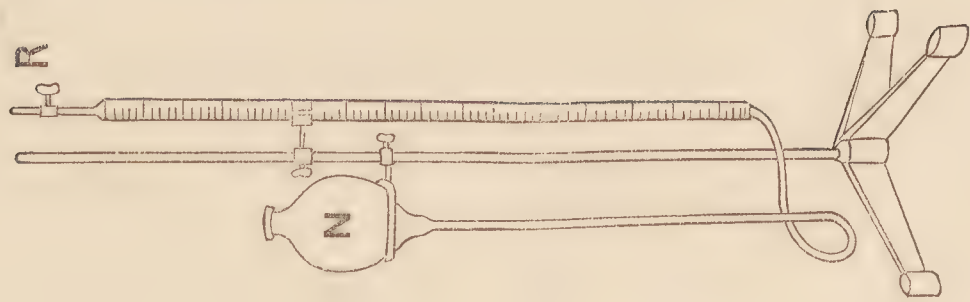


Fig. 2.

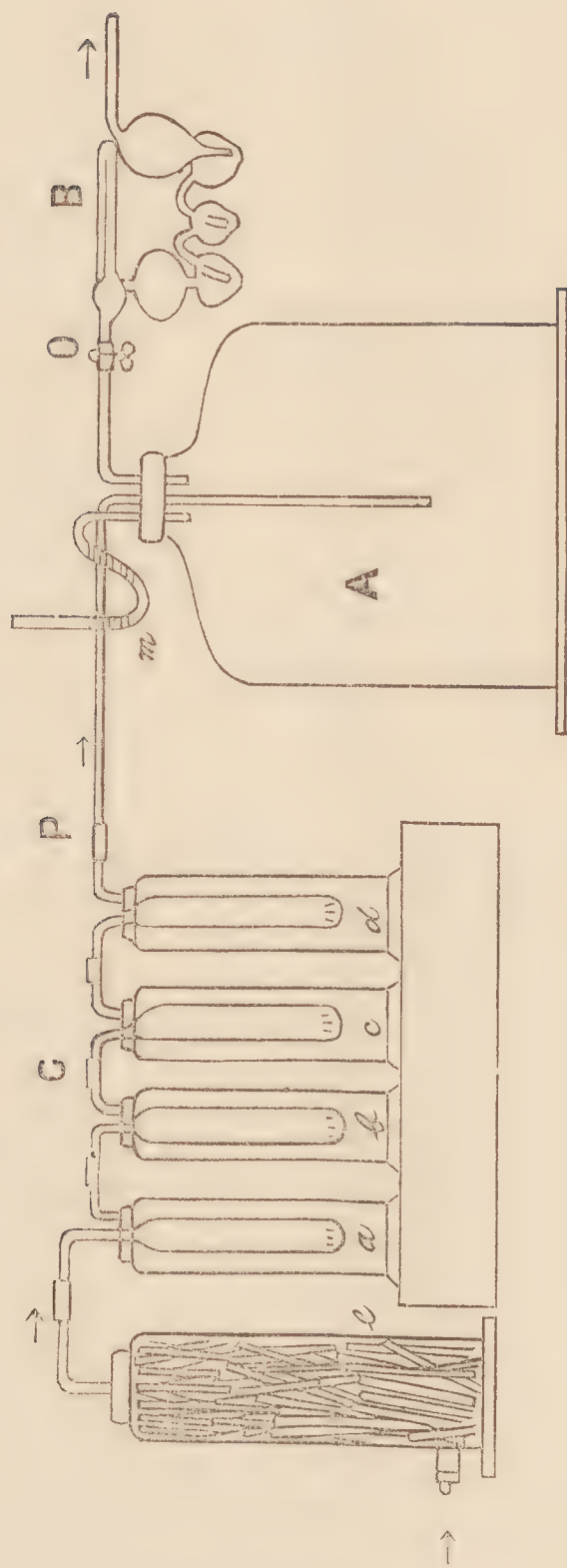


Fig. 1.

of potassium permanganate; *b* barium hydrate; *c* potassium hydrate; *d* sulphuric acid, and *e* pieces of potassium hydrate.

With the apparatus arranged as shown in Fig. 1, I determined the quantity of the carbon dioxide gas produced by a certain number of silkworms in one hour. In the belljar A, I kept 100 silkworms with some chopped mulberry leaves, air introduced into A was purified by the apparatus C, and the air in A was sucked into the potash bulb B by means of a sucking apparatus. After one hour the bulb was detached from A and weighed. In this method, the quantity of CO₂ was determined by the increase of the weight of the potash bulb. The quantity of the carbon dioxide gas thus determined was so small that it was neglected in calculations in the following experiments.

A definite volume of carbon dioxide was taken by a gas burette shown in Fig. 2. Then R (Fig. 2) was connected with P (Fig. 1), and by raising up N (Fig. 2), the gas was transferred to the belljar A; P is closed up with a pinch-cock, and O should also be shut up.

I reared worms of the race Awojiku, the same used for the first experiment (A), which hatched out on the 12th July and matured on the 6th August. The temperatures and humidities during the period were nearly the same as in the experiment (A) (*vide supra*).

In the middle of each stage the worms were placed for one hour in 0.1% and 0.5% CO₂ gas, then taken out, and reared in the same way as the control to compare with. Besides 0.1% and 0.5% carbon dioxide, 1%, 5% and 100% of the same gas were applied to the worms in the 4th and 5th ages. In the 1st, 2nd, and 3rd ages, 100 worms were used in the experiment, but only fifty in the last two ages, since their large size made it impossible to rear them in larger numbers in the jar. Beside these, fifty worms were kept in 0.5% CO₂ gas for one hour at the middle of each age.

The worms kept in 0.5% carbon dioxide soon ceased to take leaves; but those of the fourth and fifth ages looked quite indifferent to the gas. Those which were kept in pure carbon dioxide ceased to move after a minute, discharging a brown liquid from the mouth, but when taken out they revived after five minutes and took mulberry leaves like healthy ones.

The details of the above experiments were as follows:

A Control		100 worms.				
{	Ia	Worms kept in 0.1%CO ₂ for one hour, on the third day of the first stage.				
	Ib	"	"	0.5%	"	"
{	IIa	"	"	0.1%	"	" of the second stage.
	IIb	"	"	0.5%	"	"
{	IIIa	"	"	0.1%	"	" of the third stage.
	IIIb	"	"	0.5%	"	"
{	IVa	"	"	0.1%	"	" of the fourth stage.
	IVb	"	"	0.5%	"	"
{	Va	"	"	0.1%	"	on the fourth day of the fifth stage.
	Vb	"	"	0.5%	"	"
{	R	Worms kept in 9.5%CO ₂ for one hour on the third day of every stage.				
	R ¹	"	"	"	"	" except the fifth stage.
{	S	Worms kept in pure CO ₂ for two minutes on the fourth day of the fourth stage.				
	S ¹	Worms kept in pure CO ₂ for five minutes on the last day of the fifth stage.				
{	T	Worms kept in 1%CO ₂ for twenty hours at the end of the fifth stage.				
	T ¹	Worms kept in 5%CO ₂ for five hours at the end of the fifth stage.				

The results of the rearing of the silkworms in the air containing the above mentioned ratios CO₂ were as follows:

- (a) The number of diseased worm, perfect, imperfect and double cocoons per 100 are shown in the following table.

	Diseased worms before mounting.	Diseased worms after mounting.	Perfect cocoons.	Imperfect cocoons.	Double cocoons.	Worms lost during rearing.
Per 100						
A	24	8	55	4	2	5
Ia	39	4	43	4	0	10
Ib	16	0	74	0	3	4
IIa	44	8	42	2	2	0
IIb	37	3	40	2	2	14
IIIa	21	4	62	4	4	1
IIb	24	7	55	4	4	2
IVa	16	2	70	2	5	0
IVb	22	4	66	0	6	0
Va	27	8	53	4	0	8
Vb	23	12	53	0	4	4
R	20	4	60	4	4	4
R ¹	24	4	56	4	4	4
Per 10						
S	0	1	7	0	1	0
S ¹	0	0	10	0	0	0
T	4	0	6	0	0	0
T ¹	4	0	3	3	0	0

Thus no effect of CO₂ is apparent.

(b) The size of the cocoons.

	Perfect cocoon.		Double cocoon.	
	The average length of cocoons.	The average short- est diameter.	The average length of cocoons.	The average shortest diameter.
A	cm. 3.2	cm. 1.4	cm. 3.5	cm. 1.9
Ia	3.2	1.3	—	—
Ib	3.3	1.38	3.5	2.1
IIa	3.2	1.32	3.1	2.1
IIb	3.3	1.34	3.5	2.
IIIa	3.3	1.35	3.6	2.

IIIb	3.37	1.43	3.5	2.
IVa	3.2	1.31	3.4	1.95
IVb	3.39	1.34	3.2	2.03
Va	3.2	1.3	—	—
Vb	3.1	1.27	3.4	1.9
R	3.3	1.37	3.40	1.98
R ¹	3.32	1.4	3.5	1.8
S	3.3	1.3	3.5	2.2
S ¹	3.45	1.5	—	—
T	3.4	1.4	—	—
T ¹	3.46	1.47	—	—

No effect of CO₂ on the size of the cocoons can be seen.

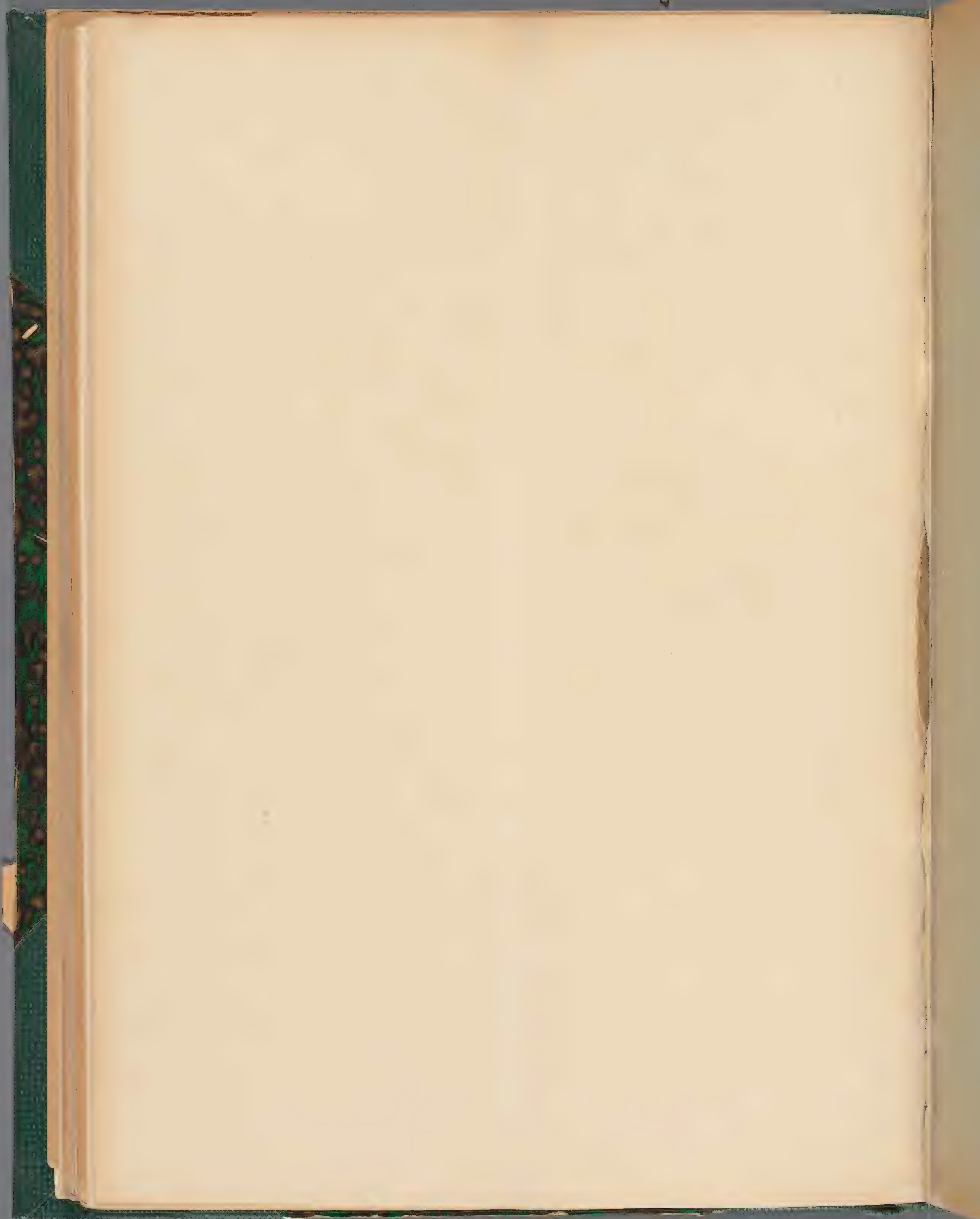
(c) The weight of the cocoons produced by worms kept in CO₂ during various periods.

(The following figures show the average weight of a cocoon).

	Cocoons with pupae.	Cocoons without pupae.
A	1.602 g.	0.171 g.
Ia	1.526	0.173
Ib	1.555	0.212
IIa	1.545	0.208
IIb	1.638	0.222
IIIa	1.598	0.212
IIIb	1.709	0.214
IVa	1.563	0.225
IVb	1.6	0.218
Va	1.292	0.175
Vb	1.373	0.232
R	1.476	0.2
R ¹	1.465	0.209
S	1.375	0.21
S ¹	1.576	0.25
T	1.59	0.212
T ¹	1.67	0.243

Conclusion.

The results of the above experiments show that the carbon dioxide, even when pure, has no influence upon the silkworms and does not act as poison, but when the worms are reared in air containing more than 5% of the gas, they lose their appetite, and their growth is more or less retarded especially in the earlier stages.



Vol. II. No. 3, PUBLISHED APRIL 30th, 1910.

Price in Tokyo Yen 0.35.

THIS JOURNAL IS ON SALE AT

Z. P. Maruya & Co., L'td.

Tori Sancho, Nihonbashi, Tokyo.

明治四十三年四月廿八日印刷
明治四十三年四月三十日發行

編纂兼發行者 東京帝國大學

印刷者

東京市麻布區市兵衛町二丁目十六番地

西谷弟松

印刷所

東京市麴町區內幸町一丁目五番地

ジャパン、タイムス社

賣捌所

東京市日本橋區通三丁目十四番地

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Vol. II. No. 4.

東京帝國大學
農科大學紀要

第二卷第四號

JOURNAL

(FORMERLY BULLETIN)

OF THE

COLLEGE OF AGRICULTURE,

IMPERIAL UNIVERSITY OF TOKYO.

TOKYO.

Published by the University.

JULY 18TH, MEIJI XLIII (1910).

PUBLISHING COMMITTEE.

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All communications relating to this Journal should be addressed to the
Director of the College of Agriculture.

Studien über die Pilze der Reispflanze in Japan.

VON

Ichiro Miyake, *Nōgakushi*.

Mit Tafel XIII und XIV.

Der Reis ist das wichtigste landwirtschaftliche Produkt in Japan, so dass reicher Ertrag oder Missernte desselben unmittelbar unser ganzes Wirtschaftsleben beeinflussen; nichtsdestoweniger hat man bisher in unserem Lande seine Krankheiten nur wenig erforscht. Daher unternahm ich es, diese zu studieren, um die wissenschaftlichen und landwirtschaftlichen Interessen zu fördern; ich erbat im Herbst 1907 von den landwirtschaftlichen Schulen und Versuchsstationen aller Provinzen sowie von gelehrten Phytopatologen die Einsendung von Exemplaren erkrankter Reispflanzen, bei welchen ich die darauf befindlichen parasitischen wie auch saprophytischen Pilze untersuchte. Da ich bei diesen Forschungen viele bisher uns unbekannte Species entdeckte, will ich hier eine kurze Monographie über die japanischen Pilze der Reispflanze geben. Da meine Studien nur kurze Zeit zurückdatieren, sind natürlich viele Lücken und Unvollkommenheiten vorhanden, welche ich später zu verbessern und zu ergänzen hoffe. Bei diesen Studien bin ich Herrn Prof. Dr. M. SHIRAI, Herrn Direktor Prof. Dr. N. MATSUI und Herrn Dr. K. HONDA zu besonderem Dank verpflichtet; der erstere bot mir freundlichen Rat und Unterweisung und stattete mich mit Literatur aus, die letzteren ermöglichten das leichte und bequeme Einsammeln der erkrankten Pflanzen. Im besonderen bin auch folgenden landwirtschaftlichen Schulen und Versuchsstationen und anderen Herren zu Dank verpflichtet:

Akita Landwirtschaftliche Schule.
Miyagi Landwirtschaftliche Versuchsstation.
Kinai Zweigstation der kaiserlichen landwirtschaftlichen Versuchsstation.
Gumma Landwirtschaftliche Schule.
Ishikawa Landwirtschaftliche Versuchsstation.
Sōma Landwirtschaftliche Schule, Fukushima.
Tottori Landwirtschaftliche Schule.
Fukushima Landwirtschaftliche Versuchsstation.
Yamagata Landwirtschaftliche Versuchsstation.
Tottori Landwirtschaftliche Versuchsstation.
Atago Forst- und Landwirtschaftliche Schule, Kyōto.
Kyōto Landwirtschaftliche Versuchsstation.
Insai Landwirtschaftliche Schule, Chiba.
Nara Landwirtschaftliche Versuchsstation.
Mie Landwirtschaftliche Versuchsstation.
Shiga Landwirtschaftliche Versuchsstation.
Inasa Landwirtschaftliche Schule, Shizuoka.
Gifu Landwirtschaftliche Versuchsstation.
Kumagai Landwirtschaftliche Schule, Saitama.
Nara Forst- und Landwirtschaftliche Schule.
Toyama Landwirtschaftliche Versuchsstation.
Ehime Landwirtschaftliche Versuchsstation.
Okayama Forst- und Landwirtschaftliche Schule.
Kagawa Forst- und Landwirtschaftliche Schule.
Ika Landwirtschaftliche Schule, Shiga.
Aichi Forst- und Landwirtschaftliche Schule.
Gotenba Landwirtschaftliche Schule, Shizuoka.
Gifu Forst- und Landwirtschaftliche Schule.
Ishikawa Landwirtschaftliche Schule.
Gumma Landwirtschaftliche Versuchsstation.
Fukuoka Landwirtschaftliche Versuchsstation.
Kagoshima Landwirtschaftliche Versuchsstation.
Okayama Landwirtschaftliche Versuchsstation.
Sagara Forst- und Landwirtschaftliche Schule, Kytōo.
Saga Landwirtschaftliche Versuchsstation.
Kumamoto Landwirtschaftliche Schule.
Rikuu Zweigstation der kaiserlichen landwirtschaftlichen Versuchsstation.
Herr KISHIRO TSUCHIYA, Tochigi.
Herr KANESUKE HARA, Gifu.
Herr KŌGORO IWAMOTO, Korea.
Herr Dr. GIKYŌ NISHIGORI, Shimane.
Herr HOLKMAN, Buitenzorg, Java.

Den Herren Prof. Dr. T. YOKOI und Prof. Dr. S. KIKKAWA bin ich für die freie Gestattung des Einsammelns der Exemplare, und Herrn II. NOMURA für die Verleihung einschlägiger Bücher aufrichtig verbunden.

Die Reispflanze ist nicht nur die wichtigste Kulturpflanze in Japan, sondern sie hat auch für die ganze Welt eine ungeheuere Bedeutung; umsomehr überrascht die Tatsache, dass die Studien über ihre Krankheiten nur vereinzelt sind; die Ursache liegt wahrscheinlich in der Schwierigkeit der Beschaffung der kranken Reispflanzen, besonders soweit Europa in Betracht kommt, wo der Reisbau lokal sehr eng begrenzt ist. Die erste wissenschaftliche Forschung in dieser Richtung wurde in Italien gemacht; in diesem Lande hat eine Krankheit, die "Brusone" genannt wird, ehemals die Reisfelder stark beschädigt, daher haben die damaligen landwirtschaftlichen Forscher und Botaniker die Ursache studiert; zugleich erschienen Beschreibungen einiger Pilze. Ein besonders bedeutungsvolles Werk war "Contributo allo Studio dei Miceti che nascono sulle Pianticelle di Riso" (Archivio del Laboratorio di Botanica Crittogamica presso la R. Università di Pavia, 1879) von ACHILLE CATTANEO, Professor an der Universität zu Pavia. Sein Werk enthält die Beschreibung von 26 Species von Pilzen, darunter waren 13 neue Species. Nach diesem erschien "Miceti del riso" von SACCARDO, der die Irrtümer CATTANEO'S berichtigte. Im Jahre 1889 publizierte der Oesterreicher FELIX VON THUEMEN eine Schrift "Die Pilze der Reispflanze," worin zwei neue Arten zur CATTANEO'schen Arbeit hinzugefügt werden. Seither ist kein eigenes Werk über die Krankheiten der Reispflanze erschienen, abgesehen von einigen Beschreibungen von Reispilzen von CAVARA, welche er seinen Studien über die italienischen Pilze einverleibt hat.

Die ersten Studien über Reiskrankheiten in Japan wurden von Prof. Dr. KOZAI im Jahre 1893 in Kyōto gemacht, und zwar über "Ishikubyō" oder Schrumpfkranheit; über die Krankheitsursache wurden seither verschiedene Theorien aufgestellt, ohne dass dieselbe bis jetzt sicher bestimmt wäre. Die "Imochi"-Krankheit der Reisflanze, die in verschiedenen

Gegenden Japans grossen Schaden verursacht und von vielen Autoren mit der italienischen Brusonekrankheit identifiziert wurde, gelangte im Jahre 1894 zum ersten Male durch Dr. K. YAMADA, Direktor der Kyōto landwirtschaftlichen Versuchsstation, zur Untersuchung. Er stellte fest, dass die Krankheitsursache ein contagiöser parasitischer Pilz ist, aber den wissenschaftlichen Namen desselben konnte er nicht präzisieren. Im gleichen Jahre hat Herr Prof. Dr. M. SHIRAI die 'Imochi' Krankheit in der Provinz Iwate studiert und sein Resultat im Amtsblatt (Kampō) No. 3785, 14. Feb. 1896 publiziert; demnach ist der Krankheitserreger sehr nahe verwandt der *Piricularia Oryzae* Br. et Cav. Andere damalige Forscher waren hinsichtlich des wissenschaftlichen Namens des Pilzes anderer Ansicht; ein Untersucher behauptete, dass derselbe zu *Cladosporium* gehöre, andere dagegen zu *Brachysporium*. Aber im Feb. 1905 hat Herr Prof. Dr. M. SHIRAI in seiner Arbeit "Supplemental Notes on the Fungus which causes the disease, so-called 'Imochibyō' of *Oryza sativa* L." (Botanical Magazine, Tokyo) den richtigen Namen festgestellt. Die erste Arbeit über die Arten der Pilze auf Reispflanzen ist "Japanese fungi of the rice-plants." von Herrn Dr. N. MIURA im Juni 1895; aber da sie nur im Manuskript vorhanden und bisher nicht publiziert ist, blieb sie leider unbekannt. In seinem Werke sind folgende Pilze enthalten:

Ustilago virens Cook.

Saccharomyces sp. (2 Arten)

Eurotium Oryzae Ahlburg.

Leptosphaeria sp.

Phoma sp. (2 Arten. Dr. MIURA reihte diese Arten der Gattung *Ascochyta* an; das kommt wahrscheinlich daher, dass er infolge des Mangels systematischer Werke falsch unterrichtet war).

Crocioreas sp.

Conicthyrium sp.

Septoria sp. (4 Arten. a. *Ascochyta*. b. und c. sind eine Species).

Hendersonia sp.

Seiridium sp.

Helminthosporium macrocarpum Grev. (=H. *Oryzae* Miyabe et Hori).

Macrosporium sp.

Sclerococcum sp.

Illosporium sp.

Botrytis sp. (2 Arten).

Vermicularia sp.

Sclerotium sp.

Undetermined fungi. (2 Arten. beide gehören zu *Dematiaceae*, *Hyphomycetes*, *Fungi Imperfecti*).

In meinen Studien konnte ich viele Arten nicht feststellen, die bereits Herr Dr. MIURA geschildert hat; aber ich hoffe sie alle in Zukunft entdecken zu können.

Ich habe in den folgenden Zeilen auch die von mir auf einer Reise im Jahre 1908 in China (Hupei, Hunan und Kiangsoo) gesammelten Pilze mit beschrieben.

ASCOMYCETES.

Gibberella Saubinetii (Mont.) Sacc.

MICH. 1, p. 513; SYLL. II, p. 554; Pilze Reispfl. p. 3; RABH. Crypt. 1², p. 102; *Botryosphaeria Saubinetii* (Mont.) Niessl., Beitr. p. 45, t. 4, f. 29; *Gibbera Saubinetii* Mont., Syll. 252 (1856); *Botryosphaeria dispersa* De Not., Sfer. ital. p. 84, t. 92 (1863); SACC. Mic. Ven. Sp. p. 118, t. 18, f. 10; Arch. Critt. 2, p. 124.

Hab. auf Blättern und Spelzen; Gumma-Landw. Schule (Oct. '07); Tochigi, leg. K. TSUCHIYA (Oct. '07).

Dieser Pilz parasitiert nach SACCARDO auf vielen Pflanzen verschiedener Gattungen und ist in Europa, Australien und Amerika verbreitet, aber es scheint, dass er keinen Schaden auf das Wachstum der Wirtspflanze verursacht. In unserem Lande wird seine Conidienform, *Fusarium roseum* Link., häufig auf verschiedenen Teilen vieler Pflanzen gefunden, aber ich habe noch nicht gehört, dass man seine Schlauchform

beobachtet hätte. Nach meinen Untersuchungen befindet sich diese auf den abgestorbenen Pflanzenteilen, wo sie mit blossen Auge als eine schwarze, verhältnismässig grosse erhabene Warze bemerkt wird, die bei schwacher Vergrösserung aus kleineren Punkten besteht, die unter dem Mikroskope als die tief blauen Perithecieen erscheinen. Die Grösse der Warze ist viel geringer auf Spelzen, dagegen ist sie viel grösser auf Blattspreiten oder Scheiden; die Zahl der Perithecieen in jeder Warze beträgt auf Spelzen zwei oder wenig mehr, während sie auf Blattspreiten oder Scheiden zehn oder noch mehr ist.

Melanomma glumarum sp. nov.

Perithecieen sind auf den Oberflächen der Spelzen zerstreut, kugelig oder ellipsoidisch, schwarz, an jedem Scheitel mit einer Mündungspapille sich öffnend, und haben einen Durchmesser von ca. $150\ \mu$; die Höhe derselben ist etwas grösser (Fig. 1.). Schläuche sind cylindrisch, sehr kurz gestielt, meist wenig eingebogen, $70-90\ \mu$ lang, $10\ \mu$ dick, 8-sporig (Fig. 2). Sporen sind zweireihig, spindelförmig, meist etwas gekrümmt, dunkel, $24-30\ \mu$ lang, $4-5\ \mu$ dick, mit drei Querwänden und in jeder Zelle sind ein bis zwei kleine Fettkörnchen vorhanden (Fig. 3.). Paraphysen werden nicht beobachtet.

Hab. auf Spelzen; Soochou, China, leg. I. MIYAKE (Dec. '08).

Die erkrankten Spelzen werden anfangs schwarzbraun verfärbt, dann allmählich gebleicht, und zugleich mit vielen Perithecieen überstreut, wobei die Reiskörner klein sind oder gar nicht zur Ausbildung gelangen; im ersten Falle sind sie braun verfärbt und daher ganz wertlos. Das Krankheitsmerkmal ist jenem der dort gewöhnlich vorkommenden durch *Phyllosticta glumarum* (Ell. et Tr.) verursachten Krankheit sehr ähnlich, demnach ist es wahrscheinlich, dass zwischen beiden Pilzen ein genetischer Zusammenhang besteht.

Mycosphaerella (Sphaerella) Oryzae (Catt.) Sacc.

Syll. I, p. 527; Mic. riso p. 11; Pilze Reispfl. p. 7; VOGL. Path. Veget. p. 145; *Pleospora Oryzae* Catt., Archiv. Trienn. 1, p.

181, t. 14-15; C. 2, p. 125; Rend. Ist. Lomb. di Sc. Ser. 2, vol 7, fasc. 5; COSSA Le Staz. Sper. Agr. vol. 3, p. 19; tav. 14-15; CANTONI Encic. Agr. Ital. vol. 1, part 1, p. 453.

Hab. auf Blättern; Shiga Landw. Vers. (Oct. '07).

Dieser Pilz erscheint nach v. THUEMEN und anderen Autoren im August oder Juli, verbreitet sich sehr rasch, und das erkrankte Blatt wird in der Regel weisslich und stirbt ab; infolge dieses Parasits ist die Entwicklung der Reiskörner sehr kümmerlich. Prof. Dr. SANTO GAROVAGLIO, der Entdecker dieses Pilzes, hielt ihn für die Ursache der Brusonekrankheit, aber nach vielen Autoren ist es bekannt, dass er häufig auf den brusonekranken Pflanzen gar nicht gefunden wurde, daher kann diese Annahme überhaupt nicht anerkannt werden; dieser Pilz verursacht in Italien und Oesterreich einen grossen Schaden. In Japan habe ich ihn nur einmal gefunden und da sein Auftreten so selten ist, dürfte sein Schaden kaum je so gross sein wie in Europa.

***Mycosphaerella (Sphaerella) Shiraiana* sp. nov.**

Perithezien sind im Gewebe eingesenkt, kugelig oder ellipsoidisch, 70-95 μ im Durchmesser und 55-80 μ in der Höhe, schwarz, nicht besonders dick, pseudoparenchymatisch (Fig. 4). Schläuche sind keulenförmig, kurzgestielt, 35-45 μ lang, 11-15 μ breit, mit acht in zwei oder in drei unregelmässigen Reihen verteilten Sporen, wobei im letzteren Falle die Sporen meist im oberen dickeren Teile dreireihig, dagegen im unteren dünneren einreihig sind (Fig. 5). Sporen sind spindelförmig, mit mehreren grossen oder kleinen Fettkörnchen versehen, 14-16 μ lang, 4,5-5,5 μ dick, zweizellig, in der Mitte eingeschnürt, indem eine Zelle etwas grösser und dicker ist (Fig. 6).

Hab. auf den beiden Seiten der Blätter, selten auf Spelzen; Atami, Izu, leg. I. MIYAKE (Aug. '06); Gumma Landw. Schule (Oct. '07); Nara F. u. L. Schule (Oct. '07); Tottori Landw. Schule (Oct. '07); Suwa Landw. Ges., Nagano, (Sept. '08); Soochou, China, leg. I. MIYAKE (Nov. '08).

Der Pilz hat viel kleinere Peritheccien, Schläuche und Sporen als *M. Malinverniana* und die Form der Schläuche und Sporen ist auch ganz verschieden. Bei *M. Oryzae* ist die Grösse der Peritheccien und Sporen von meinem Pilze nicht sehr verschieden; aber die Form der Sporen ist ganz verschieden, indem sie bei meinem Pilze in zwei ungleiche Zellen geteilt sind, während *M. Oryzae* regelmässig geteilte und in der Mitte gar nicht eingeschnürte Sporen hat. Die Schläuche sind bei diesem Pilze etwas länger und schmaler, 47-50 μ lang, 8 μ breit; endlich sind die Peritheccien bei meinem Pilze dichter zusammengehäuft als nach CATTANEO'schen Figuren. Nach meinen Untersuchungen scheint es sehr wahrscheinlich, dass der Pilz eine neue Species ist, daher nannte ich ihn *M. Shiraiana* zu Ehren des Herrn Prof. Dr. M. SHIRAI, da ich unter seiner freundlichen Anregung diese Studien in Angriff nahm.

Im August 1906 habe ich in Atami, Izu, diesen Pilz studiert, danach scheint er mir ein ganz echter Parasit zu sein; der erkrankte Teil des Blattes wird weisslich-grau verfärbt, überzieht sich mit kleinen schwärzlichen zerstreuten Flecken, deren Mehrzahl sich besonders an der Spitze oder an dem Rande des Blattes befindet; der Rand wird schwarzbraun. Unter schwacher Vergrösserung ist der Fleck eine Anhäufung von vielen Peritheccien; in den anderen, besonders neuergriffenen Partien treten reichliche Conidienträger auf, die aber regelmässig um die Flecke fehlen. Nach diesem Verhalten ist es sehr wahrscheinlich, dass die Conidienträger zu diesem Pilze gehören. Conidienträger sind einzeln oder 2-3 büschelig aus einer Spaltöffnung austretend, deren umgebende Partie etwas dicker aufgebläht ist, dunkel gefärbt mit mehreren Querwänden, gewöhnlich 40-60 μ , selten 90 μ lang, 4-5 μ dick. Der Scheitel der Conidienträger ist viel schmaler, zugespitzt und hyalin, in der Regel 10 μ , selten 25 μ lang; nach vielen Beobachtungen hat ein langer Träger einen langen hyalinen Teil, tritt aber nur vereinzelt auf (Fig. 7). Auf der Spitze des Trägers befindet sich eine Conidie, die einzellig, oval, spindelförmig oder lang-ellipsoidisch, 8,0-12,5 μ lang, 4,5-5,6 μ dick, und hyalin ist; der Inhalt derselben ist feinkörnig (Fig. 8).

Die ergriffenen Spelzen verfärben sich weisslich, die Bildung des Reiskorns wird stark gestört oder bleibt unvollständig; ich bin darum zur Annahme geneigt, dass in Japan durch diesen Parasit grosser Schaden verursacht wird.

Mycosphaerella (Sphaerella) Hondai sp. nov.

Perithechien sind im Gewebe eingesenkt, mit dünner und kurzer Mündungspapille hervorbrechend, kugelig oder ellipsoidisch, 50-60 μ im Durchmesser, selten 40 μ oder 80 μ , schwarzbraun (Fig. 9). Schläuche sind keulenförmig, kurzgestielt, gerade oder etwas eingebogen, 30-50 μ lang, 9-14 μ breit, 8-sporig (Fig. 10). Sporen sind regelmässig zweireihig oder unregelmässig dreireihig, 2-zellig, in der Mitte nicht eingeschnürt, oval, grünlich hyalin, 10-14 μ lang, 3,0-4,5 μ dick (Fig. 11). Die Zelle am dickeren Ende ist grösser aber etwas kürzer; in jüngeren Stadien sind die Sporen mit Fettkörnchen versehen aber in älteren werden diese gar nicht beobachtet.

Hab. auf Blättern; Suigen, Korea, leg. K. IWAMOTO (Oct. '07.);
Tochigi, leg. K. TSUCHIYA (Nov. '07).

Das befallene Blatt wird weisslich verfärbt und mit Perithechien überstreut, die mit blossen Auge als schwarze Punkte erscheinen. Den Pilz kann man durch die kleineren Perithechien, und die ovalen, nicht in der Mitte eingeschnürten kleinen Sporen von den anderen Species auf den Reispflanzen unterscheiden.

Sphaerulina Oryzae sp. nov.

Perithechien sind schwarzbraun, im Gewebe ganz eingesenkt, mit Mündungspapille sich öffnend, an welcher Stelle die Farbe etwas dunkler ist; kugelig oder ellipsoidisch, 65-125 μ im Durchmesser, 45-75 μ in der Höhe (Fig. 12). Schläuche sind dicht zusammenstehend, keulenförmig oder länglich, meist wenig eingebogen, 40-60 μ lang, 10-13 μ breit, 8-sporig (Fig. 13). Sporen sind zweireihig, spindelförmig, mit beiden zugespitzten Enden oft eingebogen, 4-zellig, an den Querwänden gar nicht eingeschnürt, 15-20 μ lang, 3-5 μ dick (Fig. 14).

Hab. auf Blättern; Sagara Landw. Schule, Kyōto (Oct. '07); Rikuu Zweigst. (Sept. '07); Nara Landw. Vers. (Oct. '07); Atago F. u. L. Schule (Sept. '07); Ishikawa Landw. Schule (Sept. '07); Nara F. u. L. Schule (Oct. '07); Mie Landw. Vers. (Sept. '07); Kinai Zweigst. (Oct. '07); Tochigi, leg. K. TSUCHIYA (Sept. '07); Aichi F. u. L. Schule (Oct. '07); Kāgo-shima Landw. Vers. (Oct. '07); und Shiga Landw. Vers. (Sept. '07).

Ich habe diesen Pilz nur auf abgestorbenen Blättern beobachtet, die schon weisslich verfärbt waren, daher weiss ich nicht, ob er parasitisch oder saprophytisch lebt, aber nach meinen Untersuchungen ist er in Japan sehr weit verbreitet. Unter schwacher Vergrösserung erscheint die Perithecie als ein charakteristischer schwarzer Punkt, der in der Mitte ganz schwarz ist, aber nach der Peripherie allmählich lichter wird; dadurch kann man den Pilz von anderen peritheciebildenden Pilzen im allgemeinen unterscheiden.

Phaeosphaeria gen. nov.

Perithecieen sind im Gewebe der Wirtspflanzen eingesenkt, kugelig oder ellipsoidisch, schwarz, pseudoparenchymatisch mit flacher oder warzenförmiger Mündung. Schläuche sind büschelig, 8-sporig. Sporen sind länglich mit mehr als zwei Querwänden, dunkelgefärbt. Paraphysen nicht vorhanden.

Diese von mir neu aufgestellte Gattung gehört zu *Mycosphaerellaceae*, *Sphaeriales*, *Pyrenomyces*, und unterscheidet sich von *Sphaerulina* oder *Leptosphaeria*, indem sie von jener durch die Farbe der Sporen und von dieser durch die Existenz der Paraphysen verschieden ist. Da die Farbe der Sporen und Perithecieen dunkel oder schwarz ist, nannte ich sie *Phaeosphaeria*.

Phaeosphaeria Oryzae sp. nov.

Perithecieen sind im Gewebe eingesenkt, mit Mündungspapille hervorbrechend, kugelig, oval oder ellipsoidisch, 70-125 μ im Durchmesser,

90-125 μ in der Höhe, dunkelbraun (Fig. 15). Schläuche sind cylindrisch, an der Basis etwas geschmälert, dünnwandig, 8-sporig, 35-55 μ lang, 7-9 μ dick (Fig. 16). Sporen sind zweireihig, spindelförmig, dunkelgelb, meist eingebogen, 4-zellig, an den Querswänden wenig eingeschnürt, in jeder Zelle keine oder 2-3, selten 4-5 Fettkörnchen vorhanden, 16-23 μ lang, 4-5 μ dick (Fig. 17).

Hab. auf Blättern und Spelzen. Er ist überall in Japan verbreitet.

Das erkrankte Blatt wird allmählich weisslich verbleicht, von den Rändern oder Spitzen beginnend, ohne besondere regelmässige Flecke zu bilden, bis endlich keine grüne Partie mehr gesehen wird; oft schreitet die Krankheit auch zur Scheide fort; in dieser Zeit ist das alte erkrankte Blatt wie in Fasern zerlegt und wird leicht zerbrechbar. Die kranken und gesunden Teile gehen oft direkt ineinander über, vielfach aber befindet sich zwischen ihnen ein schmaler brauner Uebergangsstreifen. Die sogenannte "Shiro-hagare-byō" (Weisskrankheit) der Reisflanze, die im westlichen Japan grossen Schaden verursacht, ist ohne Ausnahme mit dem Auftreten dieses Pilzes verknüpft. Auf den Spelzen bildet er einen schwarzbraunen Fleck, der nachher allmählich weisslich verfärbt; das Korn wird darin gar nicht oder unvollständig und braun verfärbt geformt. In jedem Falle findet sich in jüngeren Stadien *Phyllosticta Oryzae* Hori, aber allmählich erscheint mein Pilz in Mischung und endlich wird nur der letztere der einzige Parasit. Auch sind die Beobachtungen anderer Forscher mit meinen Resultaten genau übereinstimmend; daher halte ich *P. Oryzae* Hori für die Conidienform meines Pilzes.

***Phaeosphaeria Cattanei* (v. Thüm.)**

Leptosphaeria Cattanei v. Thüm., Pilze Reispfl. p. 5; Syll. IX p. 791.

VON THÜEMEN, der Entdecker dieses Pilzes, hat ihn *Leptosphaeria Cattanei* v. Thüm. zu Ehren des Prof. A. CATTANEOS genannt, und in seiner Diagnose mit dem Fragezeichen "Paraphysibus nullis (?)" beschrieben, aber in der Fussnote hat er positiv "Paraphysen wurden nicht beobachtet" festgestellt; demnach ist der Pilz nicht zu *Pleosporaceae*

gehörig, sondern muss den *Mycosphaerellaceae* eingereiht werden. Ich glaube, dass v. THUEMEN ihn falsch in *Leptosphaeria* stellt, und er richtiger in meiner neuen Gattung angeordnet werden muss.

Dieser Pilz wird von der vorigen Art durch die halbeingesenkten Peritheccien, die grossen 100-130 μ langen und 10-13 μ dicken Schläuche, die geraden Sporen und die Bildung der kleinen Flecke leicht unterschieden.

***Metasphaeria albescens* v. Thüm.**

Pilze Reispfl. p. 5; Syll. IX p. 843.

Hab. auf Blättern und Spelzen; Shingū und Hongū, Kii und Yoshino, Yamato, leg. I. MIYAKE (Aug. '06); Ika Landw. Schule, Shiga (Sept. '07); Mie Landw. Vers. (Sept. '07); Kinai Zweigst (Oct. '07); Atago F. u. L. Schule (Sept. '07); Sagara F. u. L. Schule (Oct. '07); Aichi F. u. L. Schule (Oct. '07); Kagoshima Landw. Vers. (Oct. '07); Gifu Landw. Vers. (Sept. '07); Nara Landw. Vers. (Oct. '07); Gifu, leg. K. HARA (Oct. '07).

Im August 1906 habe ich diesen Pilz, in der Nähe von Hongū, Prov. Kii, und am Ufer des Flusses Totsugawa, Prov. Yamato, entdeckt und die durch ihn hervorgerufenen Krankheitsmerkmale studiert. Nach meinen Untersuchungen bildet der Pilz einen kleinen schwarzbraunen Fleck auf dem Blatte, aber er vergrössert sich sehr rasch, gleichzeitig wird seine Farbe gelblichbraun, endlich weisslichgrau verändert, in kleinen zerstreuten schwarzen Punkten sich verbreitend. Der Pilz zerstört nicht selten alle Blätter, infolgedessen scheint er mir sogar die ganze Ernte gefährden zu können; hingegen führt v. THUEMEN in seinem Werke folgendes an: "Dass dieser Pilz die Ursache der Notreife der Samen sei, ist kaum anzunehmen, es erscheint vielmehr um Vieles wahrscheinlicher, dass er sich nur auf solchen Körnern (Spelzen ?) einfindet, beziehungsweise, dass solche einen geeigneten Nährboden für ihn abgeben, welche infolge Kränkels der ganzen Pflanze sich überhaupt nur unvollkommen ausbilden. Da der Schmarotzer einigemal in den Aehren solcher Exemplare gefunden

wurde, welche stark von *Metasphaeria Oryzae* (Catt.) Sacc. befallen und dadurch arg geschwächt waren, so spricht auch dies für eine solche Annahme. Als eigentlicher Schädling dürfte sohin diese Art nicht zu betrachten sein. Bisher wurde dieselbe nur bei Aquileja im Oesterreichischen Küstenlande im September beobachtet." Die Ansicht v. THUEMENS ist meines Erachtens nicht richtig und beruht auf unvollkommenen Beobachtungen; in meinen nachherigen Studien habe ich diesen Pilz auf Exemplaren, welche von verschiedenen Provinzen gekommen waren, gefunden; demnach ist er in Japan sehr weit verbreitet.

Dieser Pilz unterscheidet sich leicht von anderen durch die besonders charakteristischen 2-3mal verzweigten Paraphysen.

***Leptosphaeria Iwamotoi* sp. nov.**

Peritheecien sind kugelig, im Gewebe eingesenkt mit Mündungspapille hervorbrechend, dunkelbraun, 75-125 μ im Durchmesser. Schläuche sind lang cylindrisch, 55-60 μ lang, 11-13 μ breit, 8-sporig (Fig. 18). Paraphysen verhältnismässig dick aber kurz, meist nur von halber Länge der Schläuche. Sporen sind regelmässig 2-reihig, ellipsoidisch, dunkelgefärbt mit 2 Querwänden, an diesen etwas eingeschnürt, in jeder Zelle mit einem weissen feinkörnigen verhältnismässig grossen Kern versehen, 12,5-18,0 μ lang, 3,7-5,0 μ dick (Fig. 19).

Hab. auf Blättern; Suigen, Korea, leg. K. IWAMOTO (Oct. '07; Niihama, Ehime, leg. K. OBATA (Oct. '08).

Die Grösse der Peritheecien und Sporen dieses Pilzes ist nur ein Drittel der anderen Species, *L. culmifraga* (Fr.) Ces. et De Not. und *L. Salvinii* Catt., und die Zahl der Zellen einer Spore ist bei diesen Arten 8-10 und 4, während bei meinem Pilze sie regelmässig drei ist; ein besonders merkwürdiger Unterschied ist durch die kurzen Paraphysen gegeben. Ich nannte diesen Pilz *L. Iwamotoi* zu Ehren des Herrn IWAMOTO, welcher ihn zum ersten Male gesammelt hat.

***Ophiobolus Oryzae* sp. nov.**

Peritheecien sind schwarz, dick pseudoparenchymatisch, kugelig oder ellipsoidisch, im Gewebe eingesenkt, mit einer grossen warzenförmigen

Mündung sich öffnend, ca. 250 μ im Durchmesser, und ca. 300 μ in der Höhe (nebst der Mündung) (Fig. 20). Schläuche sind cylindrisch, 125-150 μ lang, 8-10 μ breit, 8-sporig (Fig. 21). Paraphysen sind fadenförmig, den Schläuchen gleichlang oder etwas länger. Sporen sind fadenförmig, eingebogen und eingedreht, 5-7 septiert, dunkelgelb, 100-130 μ lang, 2-3 μ dick (Fig. 22).

Hab. auf Blättern und Spelzen; Ishikawa Landw. Schule (Sept. '07); Kagoshima Landw. Vers. (Oct. '07); Nara Landw. Vers. (Oct. '07); Tochigi, leg. K. TSUCHIYA (Oct. '07).

Pleosphaerulina Oryzae sp. nov.

Peritheccien sind im Gewebe eingesenkt, kugelig oder ellipsoidisch, schwarz, dick pseudoparenchymatisch, mit einigen Schläuchen, 100-125 μ im Durchmesser (Fig. 23). Schläuche sind umgekehrt oval, 8-sporig, 50-60 μ lang, 30-35 μ dick (Fig. 24). Sporen sind hyalin mit 4-5 Querwänden und 1-2 Längswänden mauerförmig geteilt, 25-32 μ lang, 9-12 μ breit (Fig. 25).

Hab. auf Blättern; Ehime Landw. Vers. (Sept. '07); Gifu, leg. K. HARA (Oct. '07).

Der ergriffene Teil wird verbleicht, in demselben sind viele Peritheccien zerstreut. Weil das Auftreten dieses Pilzes selten ist, konnte ich über seine Entwicklung nicht genaue Untersuchungen nicht anstellen.

Gnomonia Oryzae sp. nov.

Peritheccien sind anfangs im Gewebe eingesenkt, dann mit langer Mündung hervorbrechend, flaschenförmig, 150-200 μ lang, 76-95 μ gewöhnlich 90-95 μ im Durchmesser, schwarz pseudoparenchymatisch (Fig. 26). Schläuche sind länglich ellipsoidisch, an dem unteren Teile etwas geschmälert, 38-45 μ lang, 12-16 μ dick, in der Regel 40×15 μ gross, 8-sporig (Fig. 27). Die Wände derselben sind dicker am unteren Teile, und dünner am oberen. Sporen sind zweireihig oder unregelmässig,

spindelförmig, gerade, 2-zellig, an den Querwänden etwas eingeschnürt, mit einigen Fettkörnchen versehen, 15-16 μ lang, 4,0-5,5 μ dick (Fig. 28). Oft ist eine Zelle der Spore etwas grösser.

Hab. auf Spelzen; Prov. Awaji, Sammler unbekannt (in unserem Institute vorhanden).

Die befallenen Spelzen sind weisslich verfärbt, woran schwarze kleine Punkte diffus sich befinden. Bei diesem Pilze ist die Verdickung des oberen Teils der Wand des Schlauches nicht zu beobachten.

FUNGI IMPERFECTI.

Bei den zu dieser grossen Gruppe gehörigen Pilzen ist, wie jeder-man weiss, nur die Conidienform bekannt, während die Schlauchform nicht festgestellt ist, daher sind die unten aufgestellten Arten zu den oben beschriebenen Ascomyceten gehörig, aber der Zusammenhang zwischen beiden Formen wird durch exakte Impfversuche konstatiert; ich will sie daher hier nach den Formen ihrer Conidien klassifizieren, und ihre Namen bestimmen. Doch stellte ich zwei Conidienformen ohne Impfversuche durch viele Beobachtungen und Erfahrungen als Ascomyceten fest; ich hoffe diese und andere Pilze in ihrem Zusammenhang weiter zu studieren.

Da man bei der Vergleichung der zu den Fungi Imperfecti gehörenden Pilzen nur Conidienträger oder Pycniden und Conidien als einzige Bestimmungsstücke benützt, so sind die Schwierigkeiten der Klassifikation viel grösser als bei den Ascomyceten, aber nach meinen vielen Untersuchungen ist es gewöhnlich, dass bei den reifen Sporen der gleichen Species, trotz der Verschiedenheit der Fundorte oder des parasitierenden Teils der Wirtspflanze, Form, Grösse und Eigenschaften der Sporen verhältnismässig konstant bleiben, ohne grosse Schwankungen, daher habe ich in diesen Studien hauptsächlich nach diesem Massstab klassifiziert.

Phyllosticta glumarum (Ell. et Tr.).

Phoma glumarum Ell. et Tr., Journ. Myc. 1888 p. 123; Syll. X. p. 185; RABII. Crypt. 1⁵. p. 337.

Diesen Pilz habe ich in unserem Lande nicht gefunden, aber er ist nach "List of Fungi on cultivated plants of Formosa" von Dr. T. KAWAKAMI und R. SUZUKI in Formosa vorhanden. Ich habe ihn an verschiedenen Orten der Provinzen Hunan und Kiangsoo, China, im October 1908 gesammelt. Der Schmarotzer parasitiert nur auf den Spelzen, indem diese dadurch anfangs schwarzbraun verfärbt, dann weisslich verbleicht und zugleich mit kleinen schwarzen Punkten überstreut werden; die Körner entwickeln sich schlecht oder gar nicht, häufig sind sie dunkelbraun verfärbt und bleiben wertlos. Ich habe ihn niemals auf Blättern gefunden.

Phyllosticta Oryzae (Cook. et Mass.).

Phoma Oryzae Cook. et Mass., Grev. XVI. p. 15; Syll. X. p. 185.

Hab. auf Scheiden; Kinai Zweigst. (Oct. '07); Atago F. u. I. Schule (Oct. '07); Miyakubo-mura, Ehime, leg. I. MIYAKE (Oct. '07).

Dieser Pilz wurde von Dr. MASSEE auf der Reisstreu von Calcutta, Indien, entdeckt, aber in Japan war er bisher unbekannt. Nach meinen Studien ist er nur auf Scheiden beschränkt, wo infolge seiner Parasitierung schwarzbraune, unregelmässige grosse Flecke gebildet werden; in extremen Fällen werden die ganzen Scheiden ergriffen, infolgedessen wird die Lebenskraft des darauf befindlichen Blattes stark geschwächt. In den Flecken sind verhältnismässig wenige Perithezien, die kugelig, dunkel bis braun und 40-50 μ im Durchmesser sind; die Sporen sind meist oval, oft ellipsoidisch oder cylindrisch mit abgerundeten Enden, 2,5-3,5 μ lang, 1,8-2,5 μ dick. Basidien sind klein.

Phyllosticta japonica sp. nov.

Pycniden sind kugelig oder ellipsoidisch, im Gewebe eingesenkt, schwarz, 75-100 μ im Durchmesser, 55-90 μ in der Höhe (Fig. 29). Basidien sind klein. Sporen sind spindelförmig, 7,5-10 μ lang, 3-4 μ dick, mit einem Fettkörnchen in jedem Pole, nicht in Ranken entleerend (Fig. 30).

Hab. auf Blättern und Spelzen; Nara F. u. L. Schule (Oct. '07).

Der Pilz ist charakterisiert durch seine spindelförmigen Sporen; die Grösse derselben ist zwischen den von *P. Oryzae* Hori und *P. necatrix*, aber von jenem unterscheidet er sich durch die Form derselben und das Entleeren in Ranken von Pycniden, von diesem kann man ihn leicht durch die ganz im Gewebe eingesenkten Pycniden abtrennen.

Das befallene Blatt wird ganz abgetötet und es erscheinen viele Pycniden auf beiden Seiten; an den Spelzen sind die Flecke wie bei vielen anderen Pilzen anfangs schwarzbraun gefärbt und dann weisslich verbleicht und mit schwarzen Pycniden überstreut. Die in jüngeren Stadien befallenen Spelzen entwickeln keine oder nur sehr kleine wertlose Reiskörner, aber die in älterer Zeit ergriffenen Spelzen sind mit braune Flecke tragenden Reiskörnern versehen; wenn diese zahlreich vorhanden sind, erscheint der ganze Reis auch bräunlich, und wird sein Wert sehr gering.

Phyllosticta Miurai sp. nov.

Pycniden sind im Gewebe eingesenkt mit Mündungspapille sich öffnend, ellipsoidisch, schwarzbraun, 80-125 μ im Durchmesser, 50-70 μ in der Höhe (Fig. 31). Basidien sind klein. Sporen sind cylindrisch mit abgerundeten Enden, 3-4 μ lang, 1,0-1,5 μ dick (Fig. 32).

Hab. auf Blättern; Kagoshima, leg. M. SHIRAI (Aug. 1892); Aichi F. u. L. Schule (Oct. '07).

Diesen Pilz hat auch Dr. N. MIURA zum ersten Male studiert, aber er hat seinen Namen nicht bestimmt, und ihn nur als *Ascochyta* sp. beschrieben. Diese Art ist charakterisiert durch die cylindrischen,

verhältnismässig langen Sporen; ich nannte ihn *P. Miurai* zu Ehren Dr. MIURAS, des ersten Beobachters.

Ausser den oben beschriebenen fünf Formen der Gattung *Phyllosticta*, gibt es noch eine andere Species, *P. necatrix* v. Thüm.; diese *Phyllosticta*-arten stellten die bisherigen Phytopathologen in die Gattung *Phoma*, aber sie parasitieren nie auf Stengeln oder Wurzeln, nur auf Blattspreiten, Scheiden und Spelzen; da die letzteren eine veränderte Form der Blätter sind, geht meine Ansicht dahin, dass sie alle zur Gattung *Phyllosticta* gehören.

Zur Bequemlichkeit habe ich hier eine Klassifikation aufgestellt.

Pycniden ganz oberflächlich	<i>P. glumarum</i>
Pycniden halb eingesenkt	<i>P. necatrix</i>
Pycniden ganz eingesenkt	
Sporen spindelförmig	<i>P. japonica</i>
Sporen cylindrisch	<i>P. Miurai</i>
Sporen ellipsoidisch	<i>P. Oryzae</i> Hori (= <i>Phaeosphaeria Oryzae</i>)
Sporen und Pycniden sehr klein	<i>P. Oryzae</i> (C. et M.)

***Chaetophoma glumarum* sp. nov.**

Pycniden sind an der Basis in dem schwarzen Hyphenfilze eingesenkt, gesellig auftretend, kugelig oder ellipsoidisch, schwarz, 80-125 μ im Durchmesser (Fig. 33, 34). Sporen sind in Ranken aus dem Munde austretend, hyalin, rundlich oval oder ellipsoidisch, an jedem Ende mit einem Fettkörnchen, 5,0-7,5 μ lang, 2,5-3 μ dick (Fig. 35).

Hab. auf Spelzen; Ochi-gun, Ehime, leg. I. MIYAKE (Oct. '07).

Die befallenen Spelzen werden weisslich verfärbt und von dem schwarzen Mycel in jeder Richtung überzogen; die Formation der Reiskörner wird dadurch stark verhindert. Auf Reispflanzen gibt es noch eine andere Species, *C. Oryzae* Cav., die man von meiner Art durch die ganz im Hyphenfilze eingesenkten Pycniden und die Grösse derselben und der Sporen leicht unterscheiden kann.

Pyrenochaeta Oryzae Shirai sp. nov.

Pycniden sind im Gewebe ganz eingesenkt, nur mit dem Mundteile hervorbrechend, schwarzbraun, ellipsoidisch, 200 μ im Durchmesser, 120 in der Höhe (Fig. 38, 39, 41). Auf dem Mundteile befinden sich μ mehrere schwärzliche, reich septierte Borsten (6-20), die in feuchtem Zustande als Mündungspapille einander gegenüberstehen, während sie in trockenem sternförmig sich öffnen (Fig. 40). Der Durchmesser des hervorbrechenden Mundteils ist ca. 40 μ , der innere desselben ca. 12 μ ; die Borsten sind 60-140 μ lang und 4-5 μ dick. Sporen sind in Ranken entleerend, spindelförmig, 4-6 μ lang, 1,5-2,0 μ dick, an beiden Enden mit einem Fettkörnchen (Fig. 42).

Hab. auf Blättern und Spelzen; Kochi, leg. Dr. S. SAWAMURA (Oct. 1895); Ika Landw. Schule (Sept. '07); Shiga Landw. Vers. (Sept. '07); Gifu Landw. Vers. (Sept. '07); Kagoshima Landw. Vers. (Oct. '07); Tochigi, leg. K. TSUCHIYA (Oct. '07).

Der von diesem Pilze ergriffene Teil wird bräunlich verfärbt und mit Pycniden überstreut; durch diese Parasitierung wird das Blatt sehr geschwächt (Fig. 36, 37).

Diesen Pilz hat im Oct. 1895 Herr Prof. Dr. S. SAWAMURA aus der Prov. Kōchi Herrn Prof. Dr. M. SHIRAI eingesandt, welcher ihn studiert und dessen Namen bestimmt, aber nicht publiziert hat. Zur Benützung seiner Studien erbat ich die Erlaubnis und füge den Pilz hier ein.

Nach Prof. Dr. S. SAWAMURAS Beobachtungen ist das Krankheitsbild wie folgt:

“Die durch diesen Pilz leicht zu schädigende Reispflanze ist gewöhnlich von mittlerer Rasse und befindet sich meist auf niederem, schwer zu entwässerndem Tonlande; die Zeit der Erscheinung der Krankheit ist vom 20. Juli bis 8. August. Das erkrankte Blatt bekommt anfangs eine gelblichweisse Spitze, aber allmählich erreicht die Verfärbung den unteren Teil und gleichzeitig bilden sich braune kleine Flecke; in dieser Zeit sind die Pflanzen sehr geschwächt. Die kranke Reispflanze wird mit einer Hand leicht ausgezogen, demnach scheint es, dass die

Wurzel auch sehr geschwächt ist. Da die erkrankte Pflanze sehr unvollständig reift, sind viele leere Samen bei der Ernte."

Sphaeronema Oryzae sp. nov.

Pycniden sind unter der Oberhaut gebildet, dann mit langem Munde hervorbrechend, flaschenförmig, 125 μ im Durchmesser, schwarz pseudoparenchymatisch (Fig. 43). Die Länge des Mundes ist ca. 100 μ . Sporen in Ranken entleerend, ellipsoidisch oder spindelförmig, 5-6 μ lang, 2,5-3,0 μ dick, hyalin (Fig. 44).

Hab. auf Spelzen; Shiga Landw. Vers. (Sept. '07).

Die befallene Partie ist weisslich verfärbt und mit kleinen Punkten überstreut.

Coniothyrium japonicum sp. nov.

Pycniden sind gesellig auftretend, im Gewebe eingesenkt, braun, an dem Mundteile etwas schwärzer, 130-150 μ im Durchmesser (Fig. 45). Sporen sind in Ranken entleerend, braun, cylindrisch oder ellipsoidisch, gerade oder eingebogen, an beiden Enden mit einem Fettkörnchen versehen, 6-9 μ lang, 2-3 μ dick (Fig. 46).

Hab. auf Blättern; Kagoshima, leg. M. SHIRAI (Oct. 1895).

Dr. MIURA machte seine Studien auf dem Exemplare, das Herr Prof. M. SHIRAI von Kagoshima gebracht hat; die Krankheit schreitet gewöhnlich von der Blattspitze oder dem Blattrand an fort, indem anfangs der Fleck braun ist, aber sich allmählich weisslich verfärbt. Der Pilz ist durch die halb-grosse Sporenlänge und -dicke von *C. Oryzae* Cav. verschieden.

Coniothyrium brevisporum sp. nov.

Pycniden sind im Flecke beisammenstehend, pseudoparenchymatisch, dunkel, ellipsoidisch, 100-130 μ im Durchmesser, 90-100 μ in der Höhe, im Gewebe eingesenkt, mit Mündungspapille hervorbrechend. Basidien sind klein (Fig. 47). Sporen sind ellipsoidisch oder oval, dunkelgefärbt, 4-5 μ lang, 2,3-3 μ dick (Fig. 48).

Hab. auf Blättern; Gumma Landw. Vers. (Sept. '07); Nara Landw. Vers. (Oct. '07); Sagara F. u. L. Schule (Oct. '07); Soochou, China, leg. I. MIYAKE (Nov. '08).

Dieser Pilz bildet einen regelmässigen oder unregelmässigen Fleck, der sich allmählich weisslich verändert, und in welchem gesellig viele schwarze Punkte sind. Von anderen Pilzen kann man ihn durch die kleinen Sporen leicht unterscheiden.

Coniothyrium anomale sp. nov.

Pycniden sind ellipsoidisch, 100-190 μ im Durchmesser, und 60-110 μ in der Höhe, dunkelbraun, im Blattgewebe eingesenkt, mit Mündungspapille hervorbrechend (Fig. 49). Basidien sind fadenförmig, verhältnismässig lang, 7,5-10 μ lang, 1 μ dick. Sporen sind grünlich braun 6-7,5 μ lang, 2-3 μ dick, ellipsoidisch oval oder spindelförmig mit ungespitzten Enden, oder oft unregelmässig; in jeder Spore ist ein weisser glänzender grosser ellipsoidischer Fettkern enthalten (Fig. 50).

Hab. auf Blättern; Awaji, Sammler unbekannt (in unserem Institute vorhanden).

Der befallene Teil, der gewöhnlich an der Blattspitze oder am Rande lokalisiert ist, wird weisslich verfärbt und der Grenzteil desselben ist schwarzbraun. Die charakteristischen Sporen und die langen Basidien unterscheiden diesen Pilz von anderen Species.

Sphaeropsis japonicum sp. nov.

Pycniden sind unter der Oberhaut, mit grosser warzenförmiger Mündung sich öffnend, schwarz, dickwandig, kugelig oder ellipsoidisch 175-200 μ im Durchmesser (Fig. 51). Basidien sind klein. Sporen sind einzellig mit mehreren kleinen Fettkörnchen, ellipsoidisch, oval, cylindrisch, oder unregelmässig, in der Reife dunkel, 12-17 μ lang, 4-6 μ dick (Fig. 52).

Hab. auf Spelzen; Ika Landw. Schule (Sept. '07).

Ein Schmarotzer, *S. Oryzae* (Catt.) Sacc., ist von meinem Pilze durch die runden Sporen leicht zu unterscheiden; der andere Pilz, *S.*

vaginarum (Catt.) Sacc., ist durch folgende zwei Punkte unterscheidbar; erstens ist seine Mündung nach CATTANEO'schen Figuren papillenförmig, klein und kurz, während mein Pilz einen grossen Mund von $40\ \mu$ Länge und $75\ \mu$ Dicke hat; zweitens ist die Form der Sporen gewöhnlich oval und die Proportion der Länge zur Breite beträgt 2:1, während bei meinem Pilze die Form derselben selten oval und das Verhältnis der Länge zur Breite 3:1 ist. Noch werden die Fettkörnchen in den Sporen, obgleich sie keinen wichtigen Unterschied bilden, in Betracht kommen; bei *S. vaginarum* ist, nach CATTANEO, der grösste Teil der Sporen mit vielen kleinen oder wenigen grossen Fettkörnchen erfüllt, dagegen sind bei meinem Pilze nur einige kleine Fettkörnchen hie und da zerstreut.

Ascochyta Oryzae Catt.

Archiv. Critt. II. p. 119; Syll. IV. p. 406; Pilze Reispfl. p. 10;

RABH. Crypt. 1⁵ p. 654; VOGL. Path. Veg. p. 235.

Hab. auf Blättern; Mie Landw. Vers. (Sept. '07); Yamagata

Landw. Vers. (Sept. '07); Tottori Landw. Schule (Oct. '07).

Dieser Pilz bildet nach v. THUEMEN keinen besonderen Fleck, auch habe ich ihn auf ganz abgestorbenen Teilen beobachtet und den Schaden durch seine Parasitierung kann ich gar nicht beurteilen.

Diplodia Oryzae sp. nov.

Pycniden sind schwarz, kugelig, unter der Oberhaut gebildet, mit Mündungspapille sich öffnend, $90\ \mu$ im Durchmesser (Fig. 53). Basidien sind klein, $3-6\ \mu$ lang, $1\ \mu$ dick, hyalin. Sporen sind ellipsoidisch, spindelförmig, oder cylindrisch mit abgerundeten Enden, russfarben, in Ranken austretend, in der Mitte mit einer Querwand, an dieser ein wenig oder gar nicht eingeschnürt, $7,5-9\ \mu$ lang, $2,5-3\ \mu$ dick (Fig. 54).

Hab. auf Blättern und Spelzen; Gifu Landw. Vers. (Sept. '07);

Gumma Landw. Schule (Sept. '07); Nara Landw. Vers. (Oct.

'07); Aichi F. u. L. Schule (Oct. '07); Tochigi, leg. K. TSUCHIYA

(Oct. '07); Awaji Sammler unbekannt (in unserem Institute vorhanden).

Diplodiella Oryzae sp. nov.

Pycniden sind oberflächlich, mit aus Hyphennetzen bestehenden Wänden, die im Querschnitte nur dünnes einreihiges Pseudoparenchym sind, kugelig oder ellipsoidisch, 120-220 μ im Durchmesser, 120-180 μ in der Höhe, dunkelbraun (Fig. 55, 56). Basidien sind klein. Sporen sind dunkel, spindelförmig mit einer Querwand, 9-13 μ lang, 2,5-3 μ dick (Fig. 57).

Hab. auf Blättern und Spelzen; Gotenba Landw. Schule (Oct. '07); Nara Landw. Vers. (Oct. '07).

Hendersonia Oryzae sp. nov.

Pycniden unter der Oberhaut mit Mündungspapille sich öffnend, braun, an dem Mundteile dunkelbraun, ellipsoidisch 100-125 μ im Durchmesser. Sporen sind in Ranken entleerend, cylindrisch mit abgerundeten Enden, braun, 10-18 μ lang, 3-4 μ dick, mit drei Querwänden, an diesen in der Reife eingeschnürt, in jeder Zelle sind 2-3 kleine Fettkörnchen vorhanden (Fig. 58).

Hab. auf Blättern und Spelzen; Fukushima Landw. Vers. (Sept. '07); Kinai Zweigst. (Oct. '07); Nara Landw. Vers. (Oct. '07); Toyama Landw. Vers. (Oct. '07).

Auf den Spelzen bildet dieser Pilz einen braunen Fleck, und für die Bildung der Reiskörner verursacht er einen grossen Schaden.

Septoria longispora sp. nov.

Pycniden sind im Gewebe eingesenkt, ellipsoidisch, mit Mündungspapille sich öffnend, 140-150 μ im Durchmesser, 100-110 μ in der Höhe, schwarz pseudoparenchymatisch. Sporen sind länglich, mit abgerundeten oder abgeschnittenen Enden, hyalin, oft eingebogen, ohne Querwand, 30-40 μ lang, 2,5-3,2 μ dick (Fig. 59).

Hab. auf Spelzen; Tottori Landw. Vers. (Oct. '07).

Die befallenen Spelzen werden grau verfärbt und die Reiskörner kommen darin nicht zur Ausbildung.

Dieser Pilz unterscheidet sich von *S. Oryzae* Catt. durch die einzelligen und zweimal grösseren Sporen; die andere Art, die auf Reispflanzen parasitiert, *S. Poae* Catt., hat nach der Diagnose weisse Pycniden, daher ist sie nach dem ENGLER'schen System nicht zu den *Sphaerioidaceae*, sondern zu den *Nectrioidaceae* gehörig und nach meiner Meinung ist sie sehr verwandt mit der Gattung *Trichocrea*, aber über die zweizelligen Sporen und ein bis dreimal verzweigten Trägern kann ich hier nichts sagen, weil ich kein Exemplar habe. Doch ist sie nicht zu *Septoria* gehörig, daher will ich sie hier bei der Vergleichung mit meinen Pilzen ausser Betracht lassen.

Septoria curvula sp. nov.

Pycniden sind ziemlich dicht zusammenstehend, kugelig oder ellipsoidisch, schwarz, 90-100 μ im Durchmesser. Sporen sind verschiedenartig eingebogen und eingedreht, mit 5-8 Querwänden und vielen Fettkörnchen, 50-80 μ lang, 2,5-3 μ dick (Fig. 60).

Hab. auf Blättern; Suwa Landw. Gesell., Nagano (Sept. '08).

Die charakteristischen Sporen unterscheiden diesen Pilz von anderen Species. Nach der Erklärung der Suwa Landw. Gesell., welche die Exemplare eingesandt hat, findet er sich auf den Reispflanzen, die durch Ueberfliessen beschädigt sind, und scheint die Krankheit infektiös zu sein; demnach gehört er wahrscheinlich zu den schwachen Parasiten.

Phaeoseptoria Oryzae sp. nov.

Pycniden sind dunkel oder schwarz, kugelig, oval oder ellipsoidisch, im Gewebe eingesenkt, dann mit Mündungspapille hervorbrechend, dick pseudoparenchymatisch, 100-150 μ im Durchmesser, 100-125 μ in der Höhe (Fig. 61, 62). Basidien sind klein, hyalin. Sporen sind in Ranken austretend, fadenförmig eingebogen und eingedreht, oft lang keulenförmig, mit abgerundeten Enden, 4-6 septiert, dunkelgelb, 30-45 μ lang, 2,5-3 μ dick, ohne Fettkörnchen (Fig. 63).

Hab. auf Blättern und Spelzen; Shiga Landw. Vers. (Sept. '07);
Sōma Landw. Schule (Sept. '07); Gumma Landw. Schule (Oct.

'07); Gotenba Landw. Schule (Oct. '07); Nara F. u. L. Schule (Oct. '07); Tottori Landw. Schule (Oct. '07); Tochigi, leg. K. TSUCHIYA (Oct. '07); Tottori Landw. Vers. (Oct. '07); Sagara F. u. L. Schule (Oct. '07); Oshima, Ehime, leg. I. MIYAKE (Oct. '07).

Ich habe diesen Pilz im Jahre 1907 entdeckt, konnte aber keine Gattung finden, der er zuzurechnen sei, daher halte ich ihn für eine Species einer neuen Gattung und nannte die Gattung *Phaeoseptoria* und die Species *P. Oryzae*, da der Pilz von *Septoria* nur durch die dunkelgelben Sporen unterschieden ist. Nachher habe ich gefunden, dass O. SPEGAZZINI in seiner Arbeit "Fungi Aliquot Paulistani" (Rev. del Huseo de la Plata 15. 1908 p. 7-48) eine neue Gattung *Phaeoseptoria* festgestellt hat; ich habe die Originalarbeit nicht gesehen, aber nach dem Referate in Annales Mycologici 1908, p. 280-281, in welchem "*Phaeoseptoria* von *Septoria* durch gefärbte Sporen verschieden" beschrieben ist, ist es zweifellos, dass seine neue Gattung mit meiner ganz übereinstimmt. Nach dem Referate seines Werkes in Hedwigia Band 48, Heft 3, p. 59-60 sind drei Species zu dieser neuen Gattung gehörig, nämlich *P. Ipirangae*, *P. Tomates* und *P. Papavae*; von diesen Beschreibungen habe ich noch nicht gewusst, aber ich glaube, dass sie von meiner Species ganz verschieden sind, da die Wirtspflanzen sehr verschieden sind, daher will ich ihn *P. Oryzae* sp. nov. nennen. Ich hoffe später diese Gattung weiter zu studieren.

Dinemasporium Oryzae sp. nov.

Pycniden sind oberflächlich mit 5-20 schwarzen Borsten umringt, umgekehrt halbkugelig, 140-160 μ im Durchmesser, schwarz (Fig. 64, 65). Basidien sind einfach fadenförmig, nur in dem unteren Teile vorhanden, 15 μ lang, 1,5 μ dick, hyalin (Fig. 66). Sporen sind ellipsoidisch oder oval, eingebogen, hyalin, an beiden Enden mit einem Haare versehen, 7,5-9 μ lang, 2,5-3,5 μ dick (Fig. 67). Die Haare sind etwas länger als die Sporen. Borsten sind gerade, dick, 2-3 mal bis 5-6 mal so lang als der Durchmesser der Pycniden.

Hab. auf Blättern; Gifu Landw. Vers. (Sept. '07).

***Dactylaria grisea* Shirai.**

Dactylaria parasitans Cav., Fung. Long. exs. III. p. 147; Syll. XI. p. 601; Bot. Mag. (Tokyo) vol. XIX. p. 24. *Piricularia grisea* (Cooke) Sacc., Mich. II. p. 148; Syll. IV. p. 217; STEV. Kans. Univ. Quart. vol. I. N. 3; *Trichothecium griseum* Cook, Rev. Amer. Fung. n. 580; *Piricularia Oryzae* Br. et Cav., Fung. Long. exs. n. 49; Syll. X. p. 563; BR. et CAV. Fung. parass. d. plant. colt. N. 188; BR. et MEN. Bol. Not. Agr. XIV. p. 672—690; Malp. XVII. p. 124—162.

Hab. auf Blattspreiten, Scheiden und Stengeln; er ist überall in Japan verbreitet.

Dieser Pilz verursacht die sogenannte "Imochi-" oder Brusonekrankheit der Reispflanze, die von älterer Zeit an in unserem Lande die Furcht der Ackerbauer ist und auch in Amerika vorkommt. In meinen Studien habe ich ihn auf von fast allen Provinzen eingesandten Exemplaren gefunden. Ueber den wissenschaftlichen Namen hat Prof. Dr. M. SHIRAI, wie oben gesagt, in "The Botanical Magazine" (vol. 19, No. 217, Feb. 1905) festgestellt, dass *Dactylaria parasitans* Cav. am richtigsten ist; aber er hat nach den "Règles internationales de la Nomenclature Botanique, adoptées par le Congrès internationale de Botanique de Vienne, 1905" den Pilz *Dactylaria grisea* Shirai genannt.

***Cladosporium Oryzae* sp. nov.**

Mycel ist oberflächlich, kriechend, indem es schwarze Flecke bildet (Fig. 68). Conidienträger, die von dort aufwärts ausspringen, sind dunkel, verschieden lang, gewöhnlich 45-70 μ lang, und 4-5 μ dick, septiert, am Scheitel mit zigzagförmigen Zähnen (Fig. 69). Sporen sind dunkel meist 2-zellig, oft 1-4 zellig, von verschiedener Grösse, 7-20 μ lang, 4-6 μ dick, an den Querwänden eingeschnürt (Fig. 70).

Hab. auf Blättern der in unserer Universität kultivierten siamesischen Reispflanze.

Dieser Pilz ist von der anderen Species, *C. maculans* Schw., durch oberflächliches Mycel verschieden,

Helminthosporium Oryzae Miyabe et Hori.

Bulletin der kaiserlichen Landwirtschaftlichen Versuchsstation No. 18. (japanisch).

Hab. auf Blättern und Spelzen; der Pilz ist überall in Japan verbreitet.

Dr. MIURA hat diesen Pilz zuerst entdeckt und hielt ihn für *H. macrocarpum* Grev., weil nach den Figuren, welche A. CATTANEO im Archiv. Critt. veröffentlicht hat, er nur durch die Länge der Sporen verschieden ist.

Cercospora Oryzae sp. nov.

Conidienträger erscheinen aus den Spaltöffnungen, einzeln oder 2-3 zusammenstehend, dunkel, mit drei bis mehreren Querwänden, 88-140 μ lang, 4-5 μ dick (Fig. 71). Am Scheitel ist die Farbe etwas dünner. Sporen sind cylindrisch oder keulenförmig, mit 3-10 Querwänden, 20-60 μ lang, 5 μ dick (Fig. 72).

Hab. auf Spelzen; Ehime Landw. Vers. (Sept. '07).

Der befallene Teil der Scheide ist braun verfärbt und mit den Conidienträgern überstreut.

Epicoccum neglectum Desm.

Ann. Sc. Nat. 17, p. 95 (1842); PENZ. in Fung. ital. tav. 127; Syll. IV. p. 737; Pilze Reispfl. p. 17; Symb. Myc. p. 373; COOKE, Handb. Brit. Fung. vol. 2, p. 560.

Hab. auf Blättern und Spelzen; Rikuu Zweigst. (Sept. '07); Ika Landw. Schule (Sept. '07); Tottori Landw. Vers. (Oct. '07); Tochigi, leg. K. TSUCHIYA (Oct. '07).

Der befallene Teil der Spelzen ist weisslich verfärbt und mit hell-schwarzen Punkten überstreut; häufig ist dieser Pilz auf den Blattspitzen der jüngeren Pflanzen.

Epicoccum hyalopes sp. nov.

Conidienträger ist hyalin kugelig, $75\ \mu$ im Durchmesser. Sporen sind glatt, schwarz, kugelig oder ellipsoidisch, $14-18\ \mu$ lang, $13-15\ \mu$ breit (Fig. 73).

Hab. auf Spelzen; Sōma Landw. Schule (Sept. '07).

Durch die charakteristischen Sporenlager kann man ihn leicht von anderen Species trennen.

Epidochium Oryzae sp. nov.

Conidienlager sind oberflächlich, warzenförmig, halbkugelig, oder unregelmässig, $160-240\ \mu$ im Durchmesser, olivenfarbig. Conidienträger sind $20-25\ \mu$ lang, $2\ \mu$ dick (Fig. 74). Conidien sind einzeln auf den Trägern stehend, spindelförmig mit zwei verhältnismässig grossen Fettkörnchen, $9-12\ \mu$ lang, $2,5-3,5\ \mu$ dick (Fig. 75).

Hab. auf Blättern; Yamagata Landw. Vers. (Sept. '07); Tottori Landw. Schule (Oct. '07); Kinai Zweigst. (Oct. '07); Kagoshima Landw. Vers. (Oct. '07); Tochigi, leg. K. TSUCHIYA (Oct. '07).
Der Pilz ist wahrscheinlich ein Saprophyt.

Sclerotium Oryzae Catt.

Archiv. Critt. 1877. p. 10; Rend. 1st. Lomb. di Sc. e Lett. vol. 9 fasc. 20; Syll. XIV p. 1153; VOGL. Path. Veg. p. 44; Pilze Reispfl. p. 18.

Hab. auf Stengeln und Scheiden; Ehime Landw. Vers. (Sept. '07); Ishikawa Landw. Schule (Sept. '07).

Dieser Pilz ist nach A. CATTANEO und anderen Autoren in Italien gewöhnlich und verursacht grossen Schaden; in Japan ist er auch an oben beschriebenen Orten vorhanden. Nach meinen und Herrn N. YANOS Beobachtungen, welcher ein Mitarbeiter der Ehime Landwirtschaftlichen Versuchsstation ist, erscheint diese Krankheit im August oder September auf den unteren Stengeln und Scheiden als schwarzbraune wolkenförmige Flecke, nachher entstehen weissliche kugelige Körnchen im Gewebe und auf demselben, die allmählich schwarz werden. In stark befallenen Partien

wird das ganze Gewebe, abgesehen von den Epidermiszellen, abgelöst und darum sehr schwach, so dass die ganze Pflanze durch das eigene Gewicht zu Boden fällt. Die obere Partie stirbt natürlich ab. Durch diese Parasitierung wird die Bildung der Reiskörner unvollständig, daher ist der Schaden sehr gross.

***Sclerotium irregulare* sp. nov.**

Sklerotien sind unregelmässig rundlich, oval, ellipsoidisch oder linear, russbraun, innen schwarz, an der oberen Seite convex, an der unteren concav, von sehr variabler Grösse 1-6 mm. lang, 1-3 mm. dick.

Hab. auf Blattscheiden; Ehime Landw. Vers. (Sept. '07); Tochigi, leg. K. TSUCHIYA (Oct. '07); Kinai Zweigst (Oct. '07); Kumagai Landw. Schule, Saitama (Sept. '07); Okayama Landw. Vers. (Sept. '07); Okayama F. u. L. Schule (Oct. '07).

Dieser Pilz unterscheidet sich von *S. Oryzae* Catt. durch die Grösse der Sklerotien und von *S. glumale* Ces. durch ihre schwarzen inneren Teile.

Diese Krankheit erscheint auf den Scheiden im August oder September; anfangs treten grünlich graue ellipsoidische Flecke auf, welche zuerst ca. 10 mm. lang und 3-4 mm. breit sind, dann vergrössern sie sich allmählich und werden graulich weiss mit schwarzbraunem Rande; im Flecke bemerkt man das farblose Mycel, welches auf oder im Inneren desselben läuft. Sklerotien, welche leicht abfällbar sind, werden gebildet zuerst auf den äusseren Seiten der Blattscheiden dann auch auf den inneren Seiten derselben. Auf Feldern, wo man Reisstreu als Dünger benutzt hat, pflegt diese Krankheit besonders häufig aufzutreten.

***Ustilaginoidea virens* (Cook.) Takahashi.**

Bot. Mag. Tōkyō, vol. 10. p. 16 (1896: *Ustilago virens* Cooke, Grev. 7. p. 15: Pilze Reispfl. p. 2; Syll. VII. p. 467; *Tilletia Oryzae* Pat., Bull. Soc. Myc. 1897 p. 124 tav. 10 fig. 2; Syll. IX p. 286; *Ustilaginoidea Oryzae* (Pat.) Bref., Unters. 1. p. 194, t. 11 fig. 22-29; Syll. XIV. p. 431.

Hab. auf Früchten; ich habe diesen Pilz überall in Japan und China gefunden.

Die BREFELD'sche Annahme, dass dieser Pilz zu den Ascomyceten gehöre, wird von vielen Untersuchern acceptiert, aber seine Schläuchform ist noch nicht entdeckt, daher habe ich ihn dieser Gruppe eingereiht.

BASIDIOMYCETES.

Tilletia horrida Takahashi.

Bot. Mag. Tōkyō, 1896. p. 20. Pl. 2; Syll. XIV. p. 422.

Hab. auf Früchten; Shiga Landw. Vers. (Sept. '07); Kagawa F. u. L. Schule (Oct. '07); Shimane, leg. Dr. G. NISHIGORI (Oct. '07).

Dieser Pilz ist in unserem Lande selten.

Bolbitius Oryzae B. et C.

N. Pac. exp. p. 62; Syll. V. p. 1077.

Hab. auf Reisstreu.

Nach den Entdeckern kommt dieser Pilz in Japan vor.

Ausser den oben beschriebenen Species gibt es noch zwei Arten, welche ich noch nicht genau bestimmen konnte; eine ist bisher als *Fusarium heterosporium* Nees. bezeichnet und unter dem japanischen Namen "Bakanae-Byō" (monströs werdende Krankheit) bekannt; aber ob der wissenschaftliche Name richtig ist oder nicht, bleibt noch dahingestellt. Die andere Art, die Dr. MIURA als *Sclerotium* sp. beschrieben hat, bildet auf Blättern längliche schwarze Flecke, die stark von diesen beschädigten Blätter sterben schliesslich ab, und das Wachstum der ganzen Pflanze wird dadurch stark gehindert. Unter dem Mikroskope haben die Flecke durch Mycel verändertes, schwärzliches Pseudoparenchym, das mit Fettkörnchen versehen ist; der äussere und mikroskopische Befund ist den Sklerotien ähnlich, aber die Lebensgeschichte des Pilzes bin ich noch studierend; ich hoffe später hierüber näheres zu veröffentlichen. Die Schrumpfkrankheit der Reispflanze, die überall in Japan verbreitet ist, bleibt in ihrer Ursache noch unbekannt.

Ich habe oben die von mir beobachteten japanischen Pilze der Reispflanze beschrieben; zur Vergleichung will ich hier die Namen der Pilze, die in andern Ländern entdeckt, aber in Japan noch nicht gefunden worden sind, anführen.

Melanospora Zamiae Corda.

Icon. I. 24. tab. VII fig. 297; Syll. II. p. 463; *Sphaeronema Zamiae* Catt., Archiv. Critt. II. p. 118. tab. XV. fig. 4; Pilze Reispfl. p. 11.

Mycosphaerella (Sphaerella) Malinverniana Catt.

Syll. I. p. 527; Archiv. Critt. II. p. 127. tab. XIV, f. 6; VOGL, Path. Veg. p. 145; Pilze Reispfl. p. 7.

Mycosphaerella (Sphaerella) Tulasnei Jaczewski.

Richerch. sur le *Cladosp. herbarum* etc. sur les cereales Crac. 1894. cum tab. (Bull. Acad. Sc. Cracovie); Syll. XI. p. 300; VOGL, Path. Veg. p. 143; *Cladosporium herbarum* (Pers.) Link, Observ. Myc. II. p. 37; Mich. II. p. 472; Syll. IV. p. 350; Pilze Reispfl. p. 14.

Metasphaeria Cattanei Sacc.

Syll. II. p. 176; Pilze Reispfl. p. 4; *Pleospora Endusiae* Fuck. var. *Major* Catt., Archiv. Critt. II. p. 125.

Metasphaeria Oryzae (Catt.) Sacc.

Syll. II. p. 180; Pilze Reispfl. p. 4; *Leptosphaeria Oryzae* Catt., Archiv. Critt. II. p. 127. tab. XIV. fig. 10.

Leptosphaeria culmifraga (Fr.) Ces. et De Not.

Syll. II. p. 75; CES. et DE NOT. Schem. Sper. p. 61; SACC. Fung. ital. t. 496; *Sphaeria culmifraga* Fr., Syst. Myc. II. p. 510; *Pleospora culmifraga* Fuck., Sym. Myc. p. 137. tab. 3, fig. 21; *Leptosphaeria culmifraga* Sacc., Myc. Ven. p. 107; Archiv. Critt. II. p. 126. tab. XV. fig. 1-9.

Leptosphaeria Salvinii Catt.

Syll. II. p. 62; Archiv. Critt. II. p. 126. tab. XV, fig. 1-3; Pilze Reispfl. p. p.

Phyllosticta necatrix (v. Thüm.)

Phoma necatrix v. Thüm., Syll. X. p. 185; Pilze Reispfl. p. 12;
RABH. Crypt. 1⁵. p. 337.

Chaetophoma Oryzae Cav.

Syll. X. p. 218; Mat. Lomb. p. 48. t. II. f. 7.; RABH. Crypt. 1⁵.
p. 449.

Coniothyrium Oryzae Cav.

Syll. X. p. 267; Mat. Lomb. p. 19

Sphaeropsis Oryzae (Catt.) Sacc.

Syll. III. p. 303; Mic. Ris. p. 4; Pilze Reispfl. p. 12; *Phoma*
Oryzae Catt., Archiv. I. p. 189. tab. XV. fig. 6. II. p. 118.

Sphaeropsis vaginarum (Catt.) Sacc.

Syll. III. p. 303; Mic. Ris. p. 4; Pilze Reispfl. p. 12; *Phoma*
vaginarum Catt., Archiv. Critt. II. p. 118.

Septoria (?) Poae Catt.

Syll. III. p. 562; Archiv. Critt. II. p. 118; Mic. Ris. p. 5; Pilze
Reispfl. p. 10; RABH. Crypt. 1⁵. p. 821; VOGL, Path. Veg. p. 238;
Dacryomyces Poae Libert., Plant. Crypt. Ard. fasc. II. n. 135.

Septoria Oryzae Catt.

Syll. III. p. 562; Archiv. Critt. II. p. 119; Pilze Reispfl. p. 11;
RABH. Crypt. 1⁵. p. 823; VOGL, Path. Veg. p. 238.

Oospora Oryzae Ferraris.

Malpighia vol. XVI. (1902.) p. 36. tab. II. 20. fig. 1—2; Syll.
XVIII. p. 498.

Papulospora sepedonioides Preuss.

Hoyersw. n. 40; Sturm. D. C. Fl. VI. t. 45; Syll. IV. 59.

Trichothecium roseum (Pers.) Link.

Syll. IV. p. 178; Observ. mycol. I. p. 16 f. 27; SACC. Fl. ital.
t. 956; Pilze Reispf. p. 15; *Trichoderma roseum*. Pers., Syn.
p. 231.

Coniosporium Oryzae (Catt.) Sacc.

Syll. IV. p. 244; Mic. Ris. p. 5. tab. XIV. fig. 11; Pilze Reispfl. p. 13.

Gymnosporium Oryzae Catt.

Archiv. Critt. II. p. 119. tab. XIV. fig. 11.

Torula graminis Desm.

Ann. Sc. Nat. 1834. II. p. 72. t. II. f. 6; Syll. IV. p. 259; Pilze Reispfl. p. 13.

Trichosporium Maydis (Gar.) Sacc.

Syll. IV. p. 293; Pilze Reispfl. p. 15; *Sporotrichum Maydis* Gar., Archiv. Critt. I. p. 39.

Monotospora Oryzae B. et Br.

Fungi of Ceylon n. 890; Syll. IV. p. 300; Pilze Reispfl. p. 13.

Cladosporium Maculans Schw.

Syll. IV. p. 355; Pilze Reispfl. p. 14; Sym. Amer. bor. n. 2599; *Helminthosporium maculans*. Catt., Archiv. Critt. II. p. 122.

Napicladium Jenseanum Rac.

Parasit. Alg. u. Pilz. Javas. II. p. 41; Syll. XVI. p. 1066.

Helminthosporium macrocarpum Grev.

Scott. t. 148; Sacc. Fl. ital. t. 825; Syll. IV. p. 412; Archiv. Critt. II. p. 121; Pilze Reispf. p. 15.

Helminthosporium sigmoideum Cav.

Mat. Lomb. p. 15. t. I. f. 5; Syll. X. p. 651.

Sphacelia Oryzae Masee.

Bull. Mic. Inform. Roy. Gard. Kew. 1899. p. 167; Syll. XIV. p. 1093.

Fusarium heterosporium Nees.

N. A. Cur. IX. p. 135; Syll. IV. p. 707; Pilze Reispfl. p. 16; Vögl, Path. Veg. p. 260; *Exosporium Lolii* Spr., Syst. IV. p. 563.

Epicoccum purpurascens Ehr.

Silv. Berol. p. 12; Syll. IV. p. 736; Pilze Reispfl. p. 17.

Sclerotium glumale Ces.

Myc. Born. p. 26; Syll. XIV. p. 1153.

Typhula filiformis (Bull.) Fr.

Esp. p. 586; Hym. Eur. p. 685; BERK. Outl. p. 285; Quél. p. 299; Clav. Bull. t. p. 187; Syll. VI. p. 749; Pilze Reispfl. p. 3.

Stropharia asota B. et C.

N. Pac. exp. n. 53; Syll. V. p. 7019.

Phytopathologisches Institut, Agrikultur-Abteilung der kaiserlichen
Universität zu Tokyo.

September 1909.

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Vol. II. No. 4, PUBLISHED JULY 18th, 1910.

Price in Tokyo Yen 0.25.

THIS JOURNAL IS ON SALE AT

Z. P. Maruya & Co., L'td.

Tori Sancho, Nihonbashi, Tokyo.

明治四十三年七月十五日印刷
明治四十三年七月十八日發行

編纂兼發行者 東京帝國大學

印刷者 西谷弟松

東京市麻布區市兵衛町二丁目十六番地

印刷所 ジャパン・タイムス社

東京市麴町區內幸町一丁目五番地

賣捌所 丸善株式會社書店

東京市日本橋區通三丁目十四番地

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Vol. II. No. 5.

東京帝國大學
農科大學紀要

第二卷第五號

JOURNAL

(FORMERLY BULLETIN)

OF THE

COLLEGE OF AGRICULTURE,

IMPERIAL UNIVERSITY OF TOKYO.

TOKYO.

Published by the University.

October 31st, MEIJI XLIII (1910).

PUBLISHING COMMITTEE.

PROF. N. MATSUI, *Ph. D.*, *Rigakuhakushi*, Director of the College (*ex officio*).

PROF. C. ISHIKAWA, *Ph. D.*, *Rigakuhakushi*.

PROF. U. SUZUKI, *Nōgaku-hakushi*.

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All communications relating to this Journal should be addressed to the
Director of the College of Agriculture.

On the Life History of *Trioza Camphorae* n. sp. of
Camphor Tree and its Injuries.

BY

Prof. C. Sasaki, *Rigakuhakushi*.

With Plates XV and XVI.

In 1907-1908, Mr. KUWAYAMA¹ described a species of Psyllidæ named *Mesohomotoma camphorae* after Prof. M. MATSUMURA, and stated that the same professor collected a large number of it at Bonin and Formosa Islands, in the latter of which it gives a great harm to the camphor trees.

The *Trioza* species, which I will describe in the following lines, is usually found on the camphor trees grown in the main island of Japan, Shikoku, Kiusiu as well as in Formosa, South China etc.

The species *Mesohomotoma camphorae*, which is said to be very injurious to camphor trees, I have unfortunately never been met with on the camphor trees in the above mentioned localities, and I am of opinion that it is never injurious to them.

The species in question, which usually swarms on the camphor trees, belongs unquestionably to the genus *Trioza* described by Messers MASKEL² and F. Löw³; but the species is quite new, and I⁴ gave it the name of "camphoræ" in 1905.

1. Transaction of the Sapporo Nat. Hist. Soc. Vol. II. Parts 1-2, 1907-1908.
2. Transaction of the New Zealand Institute Vol. XXII. 1889.
3. F. Löw: Zur Systematik der Psylloden. 1878.
4. C. SASAKI: First Report of the injurious Insects of Camphor Trees (Japanese). 1905.

***Trioza Camphorae* n. sp.**

Male: Length 1.65 mm. Body orange yellow (fig. 1). Front margin of the vertex not deeply incised. Eyes large, roundish, prominent, deep crimson red. Frontal horns (Stirnkegel) conical, bluntly ended, their ends lie apart from each other, and provided each with a clustre of short hairs. Antenna (fig. 2) short and filiform, composed of ten segments, light brownish orange with the two basal joints tinged dark brown. The two basal joints are short, broad and stout, the 3rd joint longest of all marked with about 29 ring markings, the 4th about $\frac{1}{2}$ and the 5th to 7th are each $\frac{1}{3}$ the length of the 3rd. The 8th shorter than the 7th, but somewhat thickened at the distal end. The 9th and 10th nearly equal in length, are much thicker than the 8th. The terminal or 10th segment bluntly ended and provided with a short and a long bristle. Rostrum very short, 3 segmented, the 3rd segment deep brownish black, and extends beyond the insertion of the first pair of legs. Eyes comparatively large, roundish, deep crimson red, and projected out at each side of the head. Ocelli 3, of which one lies close to each eye, and the remaining one on the middle of frons. Thorax thick and stout, and dorsally swollen. Fore wings (Fig. 3) transparent, narrow and elongated, slightly pointed at the outer margin, primary stalk of the veins more than one third the length of the whole wing; stalk of the subcosta as long as the primary stalk; radius nearly straight, reaches the costal vein at more than one fourth of the length of the wing. Cubitus bifurcating at the junction of the primary stalk of veins and the stalk of subcosta, its upper branch moderately convex at its proximal half, and again bifurcating far beyond the outer margin; the lower branch running straight for a short distance, divided again into two branchlets, the upper long and convex, and the lower short and straight. Clavus arising at the base of the wing close to the primary stalk of veins, descends and soon reaches the posterior margin. Above the clavus runs a fine delicate streak nearly parallel to the former. Hind wing (Fig. 4) transparent, and far smaller than the fore. Costal margin nearly straight, but with a

slight depression at nearly half its length. Apex rounded. A single vein arising at the base, running straight for a short distance, divided into three fine delicate branches. Upper two branches gradually diverging towards the outer margin, one reaches above and the other below the apex. Lower branch running towards the hind margin, divided into two unequal branchlets. Legs rather stout, light orange yellow, their anterior two pairs are somewhat smaller than the 3rd pair, tarsi two, claws two, lying close to the base of an expanded membranous pulvillus. The distal end of the hind tibia with three short blackish spines.

Male genitalia: Genital plate broad, as long as the genital segment, its free edge even and straight, as broad as the base, its front and hind margins are slightly constricted just beyond the two corners of the free edge. The hind margin is hollowed out lengthwise into a shallow canal. Close to the lower end of the canal lies the end of penis.

All the edges of the plate are thickly covered with fine hairs. The posterior appendages (clasper? elongated triangular, their bluntly pointed end is bent posteriorly and colored black. They are also thickly covered with fine hairs, (fig. 5). The penis (fig. 6) is composed of three segments—the 1st or basal segment is much longer than the remaining two; the 2nd is about one third the length of the first; and the 3rd is shortest, but twice as broad as the 2nd, and nearly spindle shaped. This penis, arising at the basal portion of the genital plate, descends for a short distance, and then forming a large curve ascends by passing between these two triangular appendages, and reaches beyond the base of the genital plate. If the penis is stretched out, it is about three times the height of the genital plate.

Female: Length 1.92 mm. Body larger than the male and lighter in color.

Female genitalia: The upper genital plate is rather longer than the lower, it ends bluntly, and is provided with a few long hairs. The lower, broad at the base, is also bluntly ended, and provided sparsely with hairs. The ovipositor is membranous, but it is supported lengthwise by two long chitinous spines and extends as far as the free end of the upper genital plate (fig. 7).

Deposition of Eggs.

The winged insects appear abundantly in the month of April; and less so until the month of July. The female insects lay eggs usually on the under but rarely on the upper surface of the young tender leaves of camphor trees (fig. 8). The eggs are laid in groups or not. They hatch out usually within a few days after deposition. The eggs (fig. 9) are nearly spindle shaped, one end bluntly pointed and the other, broad and round. Length 0.32 mm., breadth 0.11 mm. Egg shell rather thin, but elastic, and marked with mesh-like markings. Eggs dull yellowish grey with the broad end tinged orange yellow, but they become nearly transparent except at the broad end, just before hatching. They are tightly attached to the leaves by the broad end, and pretty difficult to detach.

Larva.

1st stage: Length 0.312 mm. Breadth 0.18 mm. Body elongated oval and flattened. The head, thorax, and abdomen are more or less distinct. The former two divisions are nearly transparent, with light yellow peripheries. Abdomen composed of seven segments, yellow, with a deep yellow triangular marking at its base. Head comparatively large, hemispherical, antenna one segmented with two hairs at its end. Eyes simple, roundish and prominent, dull yellow, and surrounded by a crimson reddish band. Filamentous mouth parts beginning just below the insertion of the first pair of legs, and forming a sort of an oval loop on the ventral surface of the head; but incapable of taking in food. Legs are stout, transparent, and appear to be composed of three segments—coxa, femur, and tibiotarsus. The end of each leg with a large pulvillus and two long hairs. The peripheries of the head and abdomen are provided with long transparent lamellar spines—each projected out from a cup-like base lying close to each other. The basal cup is orange yellow, while the spines are light orange and finely serrated at their blunt free end. The thorax with a single pair of spines—one on each side (fig. 10).

2nd stage: Length 0.444 mm. Breadth 0.288 mm. Body elongated oval, light greenish yellow. Dorsal surface flattened, ventral more or less swollen. The three divisions of the body now become distinct, head hemispherical, and fringed with a series of transparent lamellar spines. Each of the thoracic and abdominal segments with a pair of lamellar spines, the terminal segment of the abdomen with three pairs of the same. Three simple eyes, on each side of the head, light yellow, each surrounded by a broad ring of deep crimson red. In this stage, the filamentous mouth parts become unfolded, and are deeply thrust into the leaf on which the larva rests. The other characters nearly similar to those of the first stage (fig. 11).

3rd stage: Length 0.60 mm. Breadth 0.42 mm. Body oval, light greenish yellow, with an orange horse-shoe shaped marking on the dorsal surface. The dorsal surface is uniformly even, while the ventral is now more swollen than in the previous stage. The three regions of the body become more or less distinct. Head large, nearly elliptical, thorax as long as the head, but broader than the latter. Abdomen is longer but narrower than the thorax. The abdominal segments are distinct. Body is fringed with closely arranged lamellar angulated appendages. The latter are transparent, but their basal portion is dark orange. The surface of these appendages is marked with longitudinal as well as transverse fine striations. The number of the eyes, as well as the coloration of the broad band surrounding each eye, is exactly similar to that of 2nd stage. The filamentous mouth parts are deeply thrust into the tissues of leaves. Anal ring transversely elongated and lies ventrally close to the end of abdomen. The ring itself is elongated dots (fig. 12).

As the larva grows to be 0.696 mm. in length, and 0.54 mm. in breadth, the ventral surface of its body becomes more swollen than in previous stages, while the dorsal surface remains flattened. The boundary line between the head and thorax disappears so that the two form a single region—cephalo-thorax. The cephalo-thorax is much larger than

the abdomen, which is nearly hemispherical. These two regions are marked dorsally with a large horse-shoe shaped marking of dull orange color. The dorsal mid-line of the body is now marked with a straight streak, which begins at the front of the cephalo-thorax, and extends as far as the middle of the abdomen. The margin of the cephalo-thorax, where the simple eyes are located, shows a slight constriction. Each corner of the posterior margin of the same region is marked with two imperfect wing covers. The marginal cameller fringes are transparent, but their basal portion is still orange yellow. The dorsal surface is usually covered sparsely with white filamentous secretions, beside these there are six clusters of the same filaments—one on each side and in the middle of the same surface, and two close to its posterior margin. Antennae of a conical shape are marked with closely arranged transverse wrinkles, and five thicker transverse ones. Its tip ended with a single short spine. They lie wide apart from each other, close to the anterior margin of the ventral surface of the body. The basal portion of the ventral surface of the abdomen dark orange yellow. Legs stout with a single tarsus provided with a large roundish sucker, and with a single bristle. Anal ring dark orange yellow (fig. 13).

Pupa: Length 1.3 mm. Breadth 1.0 mm. Body nearly roundish, dorsally flattened, dull yellowish brown; ventrally exceedingly swollen, light orange yellow. The three regions of the body are more distinct than in the previous stage. Head large, hemispherical, and lies in a deep indentation formed at the front margin of the large and broad thorax; the wing covers developed more. Abdomen shorter and narrower than the thorax, with its posterior margin rounded. All the margins of the body with closely arranged lamellar transparent appendages similar to those in the previous stage. The dorsal surface, besides the sparse covering of white waxy filaments, bears seven groups of the latter, two groups at the anterior portion of the head; three on the thorax—one on the middle, two on the lateral sides; and two on the abdomen. On ~~the~~ dorso-median line of the body runs a distinct streak, which

begins at the front margin of the head and extends as far as half the length of the abdomen. Three simple eyes on each side of the head are now located on a round purplish red marking, which is one of the compound eyes of the future imago developed beneath the skin of the pupa. Antennae short, stout, and horn shaped, composed of two segments, the 2nd segment pointed, and its distal half marked with closely arranged ring markings. Restrum of three segments, short, bluntly pointed, with its end colored black. Wing covers larger than in the previous stage. Legs are nearly equal in size, tarsus rather long with a well developed oval pulvillus having on each edge of its base a branched claw. The pulvillus seems to form a sort of sucker. Anal ring encloses a long transverse opening, whose peripheries are covered with snowy white secretions (fig. 14).

At the time of the emergence of the imago, the skin usually splits widely open along the dorsomedian streak of the pupa.

Postembryonal Development of *Trioza camphorae* in Relation to the Formation of Galls.

The winged insects appear abundantly in the month of April, and less so in the following two months in the central provinces of our main island; but more or less earlier in the islands of Shikoku and Kiushiu as well in Formosa. At the time of their appearance, they fly about around the infested camphor trees for some days, in such numbers as to make the air cloudy. They then copulate, and the females resting on the younger tender leaves as mentioned before, lay eggs generally in groups on their under surface. The newly hatched larvæ crawl about on the same surface, and undergo soon the first moult. The larvae of the 2nd stage thrust their filamentous mouth parts deep into the tissues of leaves, and begin to suck up nourishment. At the same time, the upper surface of the leaf, below which the larva rests, is slightly elevated into a round or oval shape, thus forming an imperfect gall. The latter bears a lively greenish yellow color, and its center is decorated with a rosy or reddish dot, while its under surface is on the

contrary marked with a shallow depression, in which rests a single larva (fig. 15).

When the larva attains the 2nd stage, the galls grow 2.5-2.8 mm. in diameter, and are more swollen than in the previous stage. The galls are now colored a lively crimson red on the outside, and dull reddish purple on the inside. The surrounding regions of the gall assume a light greenish yellow coloration, which becomes conspicuous in contrast to the color of the leaves (fig. 16). In the month of June most of the larvae are in the 3rd stage, and the galls have grown larger (about 2-3 mm. in diameter). The shape of the galls remain unchanged, but their crimson red coloration becomes deeper and darker than before, the swollen ventral surface of the larvae occupies the cavity of the gall, and its opening is closed up by the flattened dorsal surface of the larvae (fig. 17).

The larvae begin to pupate from about the end of June, and continue to do so in July. At this time, the galls become greyish purple, and finally black; but their opening is still closed up by the dorsal surface provided with a certain number of clusters of white waxy filaments, so that the under surface of the leaf appears mottled with scattered white patches (fig. 18). In this condition, the pupa passes the winter, and the adult insects appear in April of the following year, and lay eggs as stated before.

The Injuries of *Trioza camphorae* to Camphor trees.

Although younger as well as older camphor trees are liable to be infested by this insect, both larva and pupa, the injuries are more serious for younger (one to ten years old) than for older trees. When the young leaves are largely infested, there may be formed large numbers of oval or roundish galls on the surface, and the growth of the leaves is retarded. Later all the infested leaves will shrink up and finally fall off. If only a few larvae become lodged on the under surface of the leaves, the latter grow to the normal size, but they may be marked either with blackish spots or patches according to the number of galls

produced by the larvae. Most of the infested leaves will sooner or later fall off and thus the growth of the trees is affected more or less. If the infestation of the younger trees is too intense, the shrunk or deformed leaves will fall off, and bring about the death of the trees.

This insect is liable to do harm to camphor trees under all sorts of cultivation, viz. to the pure or mixed camphor forests, those planted along road sides or to those grown independently far apart from each other.* The winged insects usually fly about in the neighbourhood of the localities, where they emerge, but their wings are not strong enough to enable them to fly to a distance. They are mostly carried about by winds in all directions, and when they reach camphor trees, there they lay eggs and propagate their kind. The injuries of this insect mostly do not extend higher up the tree than 9-10 feet above the ground. The trunks or branches above these limits are comparatively free from it on account of its weak power of flight.

Preventive Measures.

1. All fallen leaves, either infested or not, must be strictly gathered and burnt, or better used up for the preparation of camphor.
2. If any trees should be too much infested, cut them down without reference to their age, and use them for the preparation of camphor.
3. Whale oil, herring, shark, and sardine oil soaps of 1-2% were applied to the infested leaves, and the latter examined after 24 hours. It was found that the solutions of the whale and herring oil soaps were much more efficient than the remaining ones.

October, 1909.

Explanation of Figures

Plate XV.

Fig. 1.	<i>Trioza camphorae</i>	Male. Zeiss, A. oc. 1.
Fig. 2.	Antenna of ditto	Zeiss, D. oc. 2.
Fig. 3.	Fore wing of ditto	Zeiss, A. oc. 1.
Fig. 4.	Hind wing of ditto	Zeiss, A. oc. 1.
Fig. 5.	Male genitalia	Zeiss, B. oc. 1.
Fig. 6.	Penis	Zeiss, D. oc. 1.
Fig. 7.	Female genitalia	Zeiss, D. oc. 1.
Fig. 8.	Eggs on the under surface of the young leaves of camphor tree	Nat. size.
Fig. 9.	Eggs	Zeiss, A. oc. 1.

Plate XVI.

Fig. 10.	Larva of 1st stage	Zeiss, D. oc. 1.
Fig. 11.	" " 2nd "	Zeiss, B. oc. 1.
Fig. 12.	" " 3rd "	Zeiss, B. oc. 1.
Fig. 13.	Mature larva of 3rd stage	Zeiss, B. oc. 1.
Fig. 14.	Pupa	Zeiss, B. oc. 1.
Fig. 15.	Imperfect galls formed by newly hatched larvae	...	Nat. size.
Fig. 16.	Galls formed by the larva of 2nd stage	Nat. size.
Fig. 17.	" " " " " 3rd stage	Nat. size.
Fig. 18.	Under surface of camphor leaf occupied with many pupae	Nat. size.

Vol. II. No. 3, PUBLISHED OCT. 31st, 1910.

Price in Tokyo Yen 0.24.

THIS JOURNAL IS ON SALE AT

Z. P. Maruya & Co., L'td.

Tori Sanhome, Nihonbashi, Tokyo.

明治四十三年十月廿八日印刷
明治四十三年十月卅一日發行

編纂兼發行者 東京帝國大學

印刷者

西谷弟松

東京市麻布區市兵衛町二丁目十六番地

印刷所

シヤパン、タイムス社

東京市麹町區内幸町一丁目五番地

賣捌所

丸善株式會社書店

東京市日本橋區通三丁目十四番地

NOTICE.

VOL. I.

- No. 1, published June 16th, 1909.
No. 2, published June 26th, 1909.
No. 3, in press.

VOL. II.

- No. 1, published December 8th, 1909.
No. 2, published February 12th, 1910.
No. 3, published April 30th, 1910.
No. 4, published July 18th, 1910.
No. 5, published October 31st, 1910.
No. 6, in press.

VOL. III.

- No. 1, published August 30th, 1910.
No. 2, in press.
-

13,851

Vol. II. No. 6.

東京帝國大學
農科大學紀要

第二卷第六號

JOURNAL

(FORMERLY BULLETIN)

OF THE

COLLEGE OF AGRICULTURE,

IMPERIAL UNIVERSITY OF TOKYO.

TOKYO.

Published by the University.

A APRIL 25TH, MEIJI XLIV (1911).

PUBLISHING COMMITTEE.

PROF. Y. KOZAI, *Nōgaku-hakushi*, Director of the College (*ex officio*).

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All communications relating to this Journal should be addressed to the
Director of the College of Agriculture.

On the Chloranthy of *Prunus Mume* caused by
Caeoma Makinoi.

BY

S. Kusano.

With Plate XVII—XVIII and three Figures in the Text.

Among the numerous instances of chloranthic malformation recorded since comparatively old time (see PENZIG, '90-'94), only a very few cases are known as being caused by parasitic fungi (WIGAND, '56; GOEBEL, '84, p. 123; MAGNUS, '91; SHIRAI, '97; etc), and it appears to me that no special account has ever been given on the close relation between the fungi and the affected flowers in this peculiar class of malformations.

Some years ago I ('06) gave a brief account of *Caeoma Makinoi* and its action upon the host. The fungus infects young buds of *Prunus Mume* and causes a prominent malformation of the organs concerned. Especial attention has been directed to a typical chloranthic malformation. Since immense numbers of instances of chloranthy have already been given from numerous groups of plants, I can not be contented with giving a simple description of this malformation, but I intend to make out, if possible, the close relationship between the cause and the resulting malformation, and thus to throw more or less light upon the etiology of chloranthy. The following statements, though concerned chiefly with this point, will also comprise in part the results of pathological, teratological, and morphological studies.

The observations were made for the last few years mostly in the Botanic Garden of our College, where numerous plum-trees were seriously

affected every year and sufficient amount of fresh material for study was easily available.

Description of the Chloranthic Flower.

We shall first take a full grown affected flower and examine closely the malformations presented by each floral organ.

In the chloranth under consideration we see that each floral organ preserves its proper relative position unchanged. In a normal flower the receptacle expands, as it were, into a thin-walled cup, carrying on its margin the calyx, corolla, and androecium, the result being a perigynous flower. When diseased, it becomes much thickened and enlarged, and, becoming much shallower, carries the leafy organs in whorls (Fig. 5), as is usual in such cases.

As each phyllomorphic organ increases considerably in size, it forms altogether a large tuft of foliage leaves, which hardly reveals its derivation from a normal flower, which is very small in comparison; and the whole tuft presents a very splendid appearance when an exceedingly large amount of the yellowish-orange spores of the fungus is produced over the whole upper surface of the leaves. Such a monstrous flower is reproduced in Fig. 5, Pl. XVII in its natural colours. It will be seen at once from this figure how much nutrient substances must be drawn upon in the development of this gigantic flower.

Associated with the accelerated growth of the chief floral organs, there occurs a great increase in the length and thickness of the flower-stalk: in a normal state it is less than 2-3 mm. in length and 1.5 mm. in diameter (see Figs. 3, 4), but when diseased, it attains a length of 15-20 mm. and a breadth of 5-6 mm. (Fig. 6), and is provided with well-developed leptomal and hadromal rings surrounding a comparatively large pith.

In describing the phyllody of each floral organ, it may be noted, to avoid misapprehension, that under phyllody we mean here the substitution for the floral organs, not of the ordinary foliage leaves, but of such

diseased leaves derived from a leaf-bud, as were described in a former paper (KUSANO, '06).

CALYX (Figs. 3, 4, 6, 10).

In the normal state, the sepals are, broadly speaking, triangular, adhering to the receptacle with the broader base. As they show the least deviation from the foliage leaf, we may think *a priori* that they can be most easily metamorphosed into the normal leaves, if a necessary stimulus is given. But as a matter of fact, they appear, as far as my observations go, least liable to change, and, to speak more accurately, they assume by no means the form of a normal leaf but retain their proper form, though slightly larger and faintly serrated on the margin. Hence, in the full grown chloranthic flower, they are less easily recognisable in all cases, being concealed under the other much hypertrophied organs. The slight modification undergone by the sepals seems to be due to its being under a similar condition in relation to the parasite as the bud-scales or stipules of the lower leaves of the diseased shoot, which are also least modified. The development of the fungus is quite imperfect in all of them and no *cæomasori* are developed subsequently to the spermogonia, as is usually the case. This fact seems to lead us to the view that certain anatomical changes of the host are correlated with certain developmental conditions of the fungus, a question which will be fully discussed in a subsequent section.

COROLLA (Figs. 3, 4, 11-13).

The normal petals are nearly orbicular with a short stalk and a diameter of 10 mm. (Fig. 2). They are more subject to complete phyllody than the sepals, and the first step of their transformation into foliage leaves consists in an elongation of the stalk and a gradual expansion towards the upper portion, changing the petal into a blade with an unserrated but irregular margin, of spatulate form and light greenish colour (Fig. 3). After passing through several transitional forms the petal

becomes at length similar in shape to the diseased leaves though much smaller in size.

Frequently it happens that a certain portion, generally the apical, of some petals do not produce the caeomasori of the fungus and shows less deformation than the adjoining portions occupied by the caeomasori. The difference in the degree of the malformation between these portions is always very sharp (Fig. 13). The causal relation between the non-development of the sori and the less deformation of the organs is perhaps an important problem in the etiology of chloranthy.

ANDROECIUM.

As the stamen is much more removed in form from the original, the leaf, than the other floral organs, it is natural to expect a greater variation in its phyllomorphic change. Some stamens assume the form of a complete leaf, while others show less deviation from the original form, retaining at times rudimentary anthers or unchanged filamentous portion upon the leafy part.

The several forms thus produced from a stamen will perhaps give a basis for ascertaining the corresponding parts of a leaf and a stamen, inasmuch as a phylogenetic relation exists between the two. Keeping this matter in mind, I shall give in the following detailed account of the malformations, dividing them into six types.

1. *With double blades* (Figs. 14-16).

This anomalous stamen consists of two blades coherent, as it were, with each other by the midribs. The simplest form is shown in Fig. 14, in which the several parts of an ordinary stamen can still be distinctly seen. Here, the basal portion of the filament is slightly flattened and shows the first step in the differentiation of the surface into an upper and an under side, as may be judged from the different mode of development of the fungus on the two sides, the surface with caeomasori corresponding to the upper surface of the leaf. While the upper half of the filament remains, as the figures indicate, undeformed, though slightly enlarged,

we find on its apex, exactly in the place of the anther, four wing-like processes projecting from the portion which corresponds to the connective, two on each side, which are comparable to the front and back walls of an anther-lobe. On these processes we find the caemasori developing exclusively on the inner surface, which under ordinary conditions would develop pollen grains. This is in favour of the idea that the inner side of the anther-lobes corresponds to the upper surface of the leaf. The relative position of the hadrom and leptom in the veins of the processes also renders it highly probable.

When phyllody goes on separately in the filament and the anther, the blades derived from them become connected with each other by a narrow neck as shown in Fig. 15. This specimen differs essentially from the stamen shown in Fig. 14 in having a completely phyllomorphic filament.

In a more advanced state of change the entire stamen becomes laminar. In this case both blades are always unequal in size; so it appears as if the one accessory to the other which seems to be representative of a primary leaf (Fig. 16), seemingly coherent with their upper surface, as judged from the position of the caemasori.

Frequently it happens that the major blade has an emarginate apex and looks approximately like a dichotomous leaf which is one of the forms of phyllomorphic stamens (compare Fig. 35). It may be assumed that the double blades mentioned and the dichotomous staminal leaf have been derived by the same series of changes, but in different degrees. The dichotomous leaf would result, if the minor one of the double blades be suppressed.

2. *With a terminal filament upon the double blades* (Figs. 17, 18).

This peculiar form occurs frequently. It differs essentially from the preceding form in having a filamentous or rod-like process on the median extremity of the main blade, forming, as it were, a prolongation of its midrib. Otherwise, in the form and relative size of the two blades, as well as the manner of their connection, the present form agrees with the preceding.

The terminal filament, being slightly flattened, shows a tendency

to assume a laminar structure, producing a few caemasori on one side and thus showing a dorsoventral differentiation.

In the malformation of a stamen into a bilaminar leaf, it seems to be a general rule that the minor blade is attached to the upper portion of the major one, similarly as may be found also in the petalody of the stamen (CELAKOVSKY, '78, p. 140). Such an arrangement of four lobes on the upper portion of the leaf recalls the morphological nature of the stamen and in particular the homological relationship between the anther-lobes and the two blades.

3. *With two blades and two filaments arranged alternately*
(Figs. 19-22).

This is a much more complicated and monstrous form. Probably the change has taken place here in different degrees in the upper and lower halves of the stamen. The lower portion of the filament expands in both longitudinal and lateral directions, assuming the form of a diseased foliage-leaf, while the upper half, attached to the lower surface of that blade by the upper point of its midrib, carries again a smaller blade of irregular form, which may be supposed to have been derived from the unfolded anther-lobes. An equal expansion of each lobe produces a square plane at a right angle to the filament. Further, in the centre of this secondary blade, may arise a short filamentous process which looks like a prolonged connective (Figs. 19, 20).

Broadly speaking, the upper half of the entire staminal leaf under consideration is comparable in its mode of change and in form to an entire leaf belonging to the second form (compare Figs. 17, 18 and Figs. 19, 20). Sometimes it occurs that the upper half of the stamen is so malformed as to form a dichotomous blade which appears to be derived from the anther-lobes and which equally corresponds to some leaves of a similar form derived from entire stamens (compare Figs. 21, 22 and Figs. 35, 36).

In all the staminal leaves thus far described, we see that all the principal parts of a stamen take part in the process. In the first two forms the change has affected almost equally the upper and lower por-

tions of the stamen, while in the last the upper portion has expanded very little, giving rise to a blade which appears as an accessory to the lower one, which now becomes the main blade. At any rate, all these forms agree in that the anther-lobes have made a laminar development.

4. *With a single blade bearing a rudimentary filament*
(Figs. 23-26).

In this form only a part of the stamen partakes of the chief transformation into a leaf. The lower half of the filament constitutes the blade of various sizes and forms, and with a longer or shorter petiole, while the upper half including the anther remains unchanged, and is now found on the under surface of the blade projecting out from the midrib. The texture of this part being similar to that of a normal stamen, it soon withers and the blade then appears as if it were derived from an entire stamen.

5. *With a single blade bearing a leafy filament* (Figs. 27-29).

Closely resembling the preceding form, the lower half of the filament chiefly shows the phenomena of phyllody. While in the preceding form the forward growth of the blade went as far as to make the remaining upper abortive filament rest on its central portion, in the present form the upper half of the filament is placed immediately upon the median extremity of the emarginate blade, which exactly corresponds to the lower half of the stamen. The basal portion of the upper half of the filament is yet filamentous, while the upper expands and flattens gradually into a leaflet. Hence the whole phyllomorphic stamen appears as if it were a small petiolate leaflet derived from the extremity of an ordinary leaf by the process called "Ueberspreitung" (EICHLER, '86, p. 37, PENZIG, '02). When the change advances further, the petiolar part of the accessory blade becomes laminar and both blades are fused together (Figs. 30, 31).

6. *With a single ordinary blade* (Figs. 22-36).

The stamens undergoing the most complete phyllody may be included in this form. A single such stamen is represented by an ordinary petiolate leaf, showing no sign of its real derivation. The form of the blade is very variable. Typically it is oblanceolate or oblong resembling

a diseased leaf (KUSANO, '06, Fig. 11). Among others, of common occurrence is the form with nearly isodiametric blade and with slender long petiole, as shown in Figs. 32 and 33.

The dichotomously parted leaves are not also of infrequent occurrence, usually mixed among the leaves of the first and second forms.

Besides the staminal leaves which can be referred most naturally in the categories above enumerated, there occur intermediate or transitional forms between any two of them.

It may be added here that in a given flower all phyllomorphic stamens seem to give rise, to a certain extent, to the same form (Fig. 5), or, if not, certain form or forms occur predominantly in one and the same flower. This fact shows that in this case the stamens of the same flower, together with the other organs, are subjected equally or nearly equally to the action of the fungus.

The number of leaves appearing in one flower in place of the floral organs is very variable. Since the transformed sepals, petals, and pistil keep, so far as my observations go, to their normal number, the variation will depend upon the number of stamens undergoing the change. Even in a normal flower the number of stamens is not constant, varying from 40 to 55, but the variation is still greater in chloranthic flowers. In one case only 10 phyllomorphic stamens may be developed, while in another more than 60 may be counted. It shows that the stamens may be sometimes abortive or polyphyllic.

An examination of a large number of the diseased flowers renders it probable that there seems to exist a certain relation between the size of a full grown affected flower and the number of its phyllomorphic organs. When the number of leaves is less, their development is vigorous, becoming larger and thicker than when they are more numerous. This relation may depend to a certain extent upon the nutritive condition of the flower-bud, and it is evident that the same quantity of nutritive substances will induce, other things being equal, a better development of the floral organs

when these are less numerous. Generally speaking, the fact that the diseased shoot and flower are somewhat smaller when they occur together in a common axil than when they stand alone, each in different axils. is in support for this view. Further evidence in the same direction may be found in a flower with a gigantic carpellary leaf, represented in Fig. 53, where the floral organs are mostly suppressed.

To show the relation of what have been stated in the last three paragraphs I shall produce a table showing the size of the flower, and the form and number of the stamens developed:

Size of flower. ¹	Number of stamens developed.	Type of phyllody.	Remarks.
Moderate	10	All 4	With short petiole and cleft blade.
Small	10	4, rarely 3	Half the whorls undeveloped; blade greatly cleft.
Moderate	11	All 4	Blade long, cleft; half the whorls unchanged.
Large	16	All 4	Ordinary leaf with oblanceolate form; pistil prolific.
Large	16	All 4	Stalk short; blade cleft and short; together with diseased shoot; with many staminal leaves not completely developed.
Large	18	All 4	With oblong or lanceolate blade.
Small	25	All 4	Blade small; the peripheral ones cleft, the interior oblong.
Smallest	25	1, 3, 5	Together with shoot; with long filamentous portion in the phyllomorphic anther.
Moderate	23	4	
Small	30	All 4	With large pistillary leaf.
Moderate	30	All 4	Petiolate blade cleft star-like.
Moderate	30	4	
Small	32	4, 5	
Moderate	35	3	
Moderate	35	2, some 5	
Large	35	Mostly 4	With long petiole and cleft blade.

1. By "large" we mean the size as shown in Fig. 5.

Size of flower.	Number of stamens developed.	Type of phyllody.	Remarks.
Moderate	35	4, rarely 3	Blade long, irregularly parted; mixed with small leaves; with seven unchanged stamens.
Small	35	4, rarely 5	Blade lanceolate, stipulate.
Large	37	All 4	Blade cleft and with long petiole.
Small	37	3, 4 or 5	
Large	40	4, 5, 6	
Small	40	All 5	Blade palmate.
Moderate	40	All 4	Blade cleft and with long petiole.
Moderate	40	Mostly 4, some 3	Blade cleft and with long petiole.
Small	40	4, 5	
Large	44	All 4	Together with leaf-bud.
Moderate	44	All 4	Together with leaf-bud.
Large	45	All 4	Blade cleft and with long petiole.
Moderate	45	Mostly 5, rarely 4 and 3	Together with leaf-bud; with long petiole.
Moderate	45	4 and 5, rarely 1	Together with leaf-bud; blade lanceolate.
Small	47	1, 4, mostly 5	
Moderate	50	4, 5	Blade cleft and with long petiole.
Small	50	5, 6	Blade of 6. type lanceolate.
Moderate	52	All 4	Blade cleft and with long petiole.
Large	55	All 4	Blade cleft and with long petiole.
Smallest	55	All 6	Approximately like ordinary leaf with short petiole.
Moderate	60	All 4	Blade petiolate, with stellate clefts.
Moderate	60	4, few 5	Blade isodiametric and with long slender petiole.
Smallest	62	All 4	Like ordinary leaf, oblong in form.
Moderate	70	All 4	Like ordinary leaf.
Smallest	86	All 4	Blade oblanceolate, prolifical.
Small	?	1, 2	

GYNAECIUM (Figs. 51-54).

Generally, the pistil is more liable to show the phenomena of phyllody than the stamen. So in the chloranthic flower of *Prunus*, it undergoes, first of all, a complete change even when the other organs, such as stamens and petals, are somewhat incompletely phyllomorphic. Yet, it may be often the case that the style, when the caemasori do not appear on it, retains its original shape and character, remaining unchanged on the top of the blade and thus corresponding, as it were, to the tapering apex of a normal leaf. The form of the derived leaf is more or less variable; mostly it is, besides being oblong like a normal leaf, trilobate (Fig. 54), or irregularly cleft, always with a long petiole. Of the lobes of the blade it is a noteworthy fact that the lowest or lower pair attains the largest size, while the others are small (Figs. 52, 53). As a rule, the leaf derived from the pistil is the largest of all transformed organs of a single flower.

A somewhat incompletely phyllomorphic pistil is shown in Fig. 51. On its margin two ovules are represented in an incomplete stage of phyllody; the one on the left (*a*) is a blade rolled up and the other on the right (*b*) a blade parted in two lobes. On the centre of the upper surface of both blades may be seen small papillae representing the nucelli of the ovules.

In connection with chloranthy another related malformation occurs frequently on the androecium and gynaecium, and we have, instead of leaves, shoots with a short axis and few leaves, representing the proliferation of flowers. Such a change is most likely to occur on those flowers that have undergone the most complete phyllody, and therefore it appears that the proliferation represents a more advanced stage of malformation due to the parasitic fungus (Fig. 5).

In the diseased flower, in which we have counted 86 oblanceolate, small, nearly perfect leaves in the place of stamens (see Table given above), are developed a few short shoots in the whorl of the androecium, each

carrying two or three oval, stipulate leaflets (Figs. 41. 42). As they arise from the axils of the stamens, the term 'axillary proliferation' may fitly be applied to this phenomenon. A similar change takes place more frequently in the gynaeceum, and in this case the term 'median proliferation' may be used (Fig. 5). The growth of the shoot is here far more vigorous than in the case of the androecium.

It may be interesting from the etiological point of view that the proliferation is always associated with the most complete chloranth. Since we know that the malformation under consideration is due to the action of a fungus, it may be reasonably supposed that in this case the fungus acts upon the flower at an earlier stage, probably prior to the completion of the embryonal stamen or pistil.

Development of the Diseased Flower-Bud.

It must not be supposed that all diseased flowers attain such a full development as is described in the foregoing: some of them wither out sooner or later after blooming. To make clear the influence of the various degrees of affection on the fate of the flowers concerned, how far the change can advance in young flowers, and again to get a knowledge of the ontogenetic development of each phyllomorphic organ, we will now proceed to study the diseased flowers in the younger stages of their development. The first question is that of the symptom of the disease first visible in the bud.

At the end of January, the diseased flower-bud shows no external signs of abnormality, being still at rest, small, and compact. Generally, the first indication of abnormality becomes apparent at the time the bud begins to swell up, and the greenish corolla comes in sight. On other points, for instance, the rate of the unfolding of the bud, the time of blooming, and its subsequent growth, the difference between the normal and abnormal flower-buds is not evident. In Figs. 2-4 are represented slightly affected and completely deformed flowers at the beginning of April. The normal flowers have just withered, while the partially affected (Fig.

2), incompletely (Fig. 3) and completely (Fig. 4) chloranthic flowers are nearly like normal ones in size and general form. At this stage the diseased flowers are distinguished from the normal by their pale yellowish colour and the thick texture of the floral organs. The diseased leaf-bud is somewhat accelerated in development: it is in a stage corresponding to that of the diseased flower, while the normal buds remain still closed (compare Fig. 1 and Fig. 3). It is clear from this fact that the gigantic size of the full grown chloranthic flower is due to continued growth till a later period, proceeding at an equal pace with the normal or diseased shoot.

To what degrees the malformation will proceed in a given flower can be told already at the flowering stage or still earlier, so that all the affected flowers can be confidently classified into the three groups, according to the degrees of malformation which they already manifest.

1. Partially affected flower (Fig. 2). It differs from the normal flower to a very slight degree. Only the basal portion of some or all petals and stamens develop the spermogonia. The affected parts are yellowish or pale greenish in colour, and are slightly elongated and thickened, while the healthy parts present the normal appearance. More remarkable is the change of the pistil, which, without losing its general form, becomes elongated and swollen, and is always more hypertrophied than the other organs. It may be noted that in a partially affected flower the organs of the inner whorls are more frequently and more extensively attacked by the fungus than those of the outer. None of the organs of such a flower can develop into a greenish leaf.

2. Incompletely chloranthic flower (Fig. 3). The petals and pistil undergo complete phyllody. But the stamens are not completely deformed: only the basal portion of the filament becomes a narrow blade with serrate margin, and the upper portion, developing quite normally, carries at times a perfect anther containing pollen grains, though in most cases the anther is sterile and greenish, and the connective has become broad. The stamen at this stage is nearly as large as the normal one. The pistil shows separation along the suture line, the hairs covering its whole surface in the normal state are suppressed in development (Figs.

43, 44), and the whole organ looks like a young, rolled-up leaflet (Figs. 4, 49). The apex of such a pistil sometimes bears an imperfect stigma. The spermogonia are found scattered over the whole surface of the floral organ.

3. Completely chloranthic flower (Fig. 4). All the organs are replaced by foliage-leaves. In the stamen the filament and anther become laminar separately or together. Before the unfolding of the bud all the organs look like bud-scales, with the exception of the pistil which projects from the centre of the bud as a young leaf twisted and rolled up. Spermogonia are found on all the organs.

The flowers of the first group, having least deviated from the normal flower, wither soon after blooming. The flowers belonging to the other two groups persist in their abnormality, and can grow further for a considerable length of time (Fig. 5). Thus, examination of the young flowers show that the form which a malformed stamen will assume when full grown, though very complex, is already determined at the flowering stage.

In comparing some partially phyllomorphic organs, such as stamens, or pistils, in the young and adult stages of growth, we notice that the undeformed part of each organ, generally the upper, does not show any noticeable increase of size or other change during the growing period (Figs. 3, 23-26, 49, 51-53). The extraordinary hypertrophy of the most completely phyllomorphic organs is mainly due to the abnormal enlargement of their basal portion which is known to remain meristematic, i.e. capable of cell multiplication, longer than the apical portion. The several forms shown in Figs. 19-29 will confirm this statement.

It follows that the local enlargement of organs takes place chiefly in an incompletely chloranthic flower; but, in general, in the most completely chloranthic flowers, such as is shown in Fig. 4, the organs show a tendency to enlarge uniformly, so that in both the younger and adult stages the form of the leaves is nearly similar, being lanceolate or oblanceolate.

Thus, having distinguished two cases of the enlargement of organs

according as they are in the state of complete or incomplete phyllody, we are inclined to think that the chief cause of this difference is perhaps due to the different action of the fungus according to the time of infection. The uniform enlargement would be hardly possible in those organs, in which the most important differentiation of tissues has been nearly completed, and no or very little meristematic portion is left. If, on the other hand, the fungus should infect the organs at an earlier undifferentiated stage, the mycelium would be able to extend uniformly through the entire organ and give rise to a uniform deformation.

Relation between the Development of the Fungus and the Malformation of the Floral Organs.

Comparison of the diseased organs in young and adult stages leads us to the view that the change brought about by the fungus is different in degree according as the latter develops only spermogonia or both spermogonia and caeomasori. The affected organ already shows a certain degree of malformation at the time the spermogonia appear: the formation of chlorophyll begins, serration is usually found on the margin, and more or less hypertrophy is taking place. At this time the size of the flower is nearly as shown in Fig. 3 and 4. The further enlargement of the organ accompanied by other changes takes place when the fungus has developed so far as to produce the caeomasori, and it is at the time of maturation of the caeomasori that the organ attains full growth. Moreover, it may be observed that, where the sori do not appear, the malformation does not proceed further, but remains in the state in which it was when the spermogonia appeared. Even in full grown chloranthic flowers we invariably see the clearest difference in the degree of malformation between the organs, or portions of an organ, with and without caeomasori. The very slight malformation not unfrequently met with in sepals and some bud-scales, which we have already mentioned, must certainly be closely related with this circumstance of the development of the fungus. The irregularly shaped phyllomorphic petals, such as are shown in Figs. 11-13 seem to bring out this relation more clearly. The portion occupi-

ed by the spermogonia is thin, and often has a reddish tint with less chlorophyll, while that occupied subsequently by the caeomasori is very thick and pale greenish. These characters are sharply demarcated from each other on the boundary line between the two portions. In the figures mentioned, the portion occupied by the caeomasori appears to have developed mainly after the maturation of the spermogonia, showing secondary growth. The asymmetrical forms shown in Fig. 11 and 13 are certainly due to such growth having taken place unequally on both sides of the basal portion, while a uniform growth of this portion would result in a symmetrical form (Fig. 12).

A similar relation is also found in the pistil. Caeomasori are sometimes absent on the style, and the latter is then but slightly modified, retaining sometimes its original form and bearing a stigma. To the same cause is also due the frequent presence of undeformed filaments on the leafy portion of a transformed stamen (Figs. 23-26).

It still remains to inquire into the causal relation between the different degrees of malformation and the mode of development of the fungus. Is the condition of the fungus itself leading to the production of caeomasori the direct cause of the advanced malformation, or is the production of caeomasori correlated with such internal condition of the host as to bring about advanced malformation? In my opinion, the chloranthic malformation in question is doubtlessly due to the action of the fungus, but the degree of malformation may be determined by the internal condition of the affected organ itself. While it is believed that the production of the caeomasori is a proof of the vigorous development of the fungus, for which the affected organ must provide a sufficient amount of nutritive substances, it is quite certain that the fungus would be unable to produce the sori, were the organ not responsive to this new condition of the fungus. A great hypertrophy of the organ or its part, which is always associated in the present case with the more advanced malformation, shows that it is capable of the multiplication of cells. When we remember the fact that the upper portion of an organ is less subjected to malformation than the basal portion where the multiplication of cells and the consequent

hypertrophy as well as the production of caeomasori can take place more easily than in the former, the malformation seems to depend directly on the state of the organ or its portion: while the basal portion is still meristematic, the fungus can irritate it by its development so as to cause hypertrophy. In short, the development or non-development of the caeomasori must be ascribed to the condition of the affected portion and the same condition gives rise to different degrees of malformation.

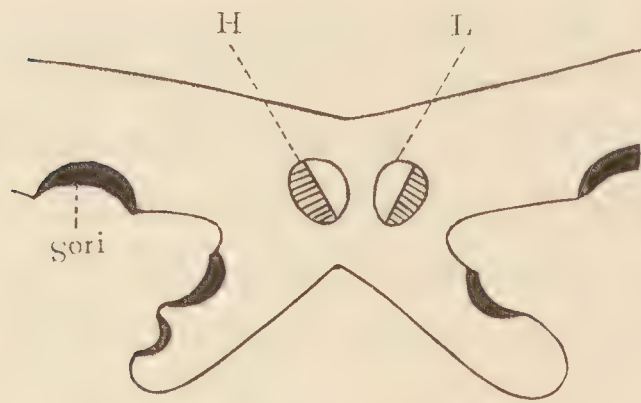
Malformation of the Anther.

From the chloranthic flowers we have obtained several forms of deformation in the upper portion of the staminal leaf. The gradual transition from one form to another expresses, in my opinion, the different degrees of abnormal development of the anther. Believing this direction of the abnormal development of the anther to be parallel to that of the phyllomorphic malformation of other organs, it may be proposed that the several forms presented by the anther afford some evidences for its morphological value, assuming that teratological data may be made a basis for morphological study¹. As to the morphological value of the anther several conflicting views are held at present. This conflict of opinions is chiefly due to a different valuation by different writers of the chief evidences on which their views are based. From the historical review of MASTERS ('69) and CELAKOVSKY ('78) we see that some older writers believed the loculi of the anther to have been derived by excavations of the leaf parenchyma, at the marginal portion, on both sides of the blade, two on the upper and two on the under surface, while the margin itself became the suture of the anther; while some botanists hold the view that the connective corresponds to the median portion of the staminal leaf, the loculi being derived from the lateral portion of lamina, by a diminution of its length and breadth, and an increase of its thickness. A. BRAUN'S ('76) opinion as to the formation of the anther, afterwards sup-

1. I leave the discussion of this fundamental point to such recent writers as GOEBEL ('98 p. 153) and VELENOVSKY ('05, p. 15), who hold opposing views.

ported by WYDLER, is that the blade becomes 4-winged by enation, the upper two wings being newly enated, while the lower two are parts of the primary blade, and each two wings on the two sides form one half of the anther. On the other hand, SACHS ('82, p. 541) holds a quite different view that the parts of the stamen corresponding to a leaf are the connective and the filament, and the anther-lobes are produced in some cases at least secondarily as an appendage. Of recent writers CELAKOVSKY ('78, '84) adopts BRAUN's theory, ENGLER ('75) and VELENOVSKY ('10, p. 939) agree with the older botanists, while GOEBEL follows SACHS.

Taking into consideration the facts obtained by the present study, we are inclined to the view of the "Verdoppelung" of the anther, and are strongly in favour of BRAUN's theory. In the phyllomorphic stamens shown in Figs. 14, 19-22, we find a leafy structure in the place of the anther, which I assume to represent a metamorphosed anther. Likewise I am of the opinion that the bilaminar structure at the apex of some phyllomorphic stamens (Figs. 15-18) is produced by an abnormal development of the anther. Bringing these structures together we may perhaps conclude that each two wing-like processes of the bilaminar leaf exactly correspond to each anther-lobe, as BRAUN mentioned. Fig. 14, which shows a slight degree of phyllody, will amply justify this statement. Also there is no doubt that the small four-lobed blade in Figs. 19, 20 represents an anther or anther-lobes. If the phyllomorphic change goes simultaneously in the filament and anther, a bilaminar leaf would result (Figs. 15-18). Taking together such bilaminar leaf and the dichotomous leaf (Figs. 35, 36) found in the most completely chloranthic flowers, it appears that there is a close genetic relation between these two forms. The minor blade in the bilaminar leaf corresponds to the inner halves of the two lobes in the dichotomous leaf, the former being formed from the latter by the bulging out towards the upper surface. The evidence in favour of this view and against the view that the bilaminar structure is formed from two different leaves, minor and major, fused together along the midribs will be found in the arrangement of the bundles at the junction point of the four wing-like processes, as well as the dis-



Text-fig. 1.

tribution of the veinlets in the latter. In a cross-section through the bilaminar portion we find two vascular bundles at the junction point. As shown in Text-fig. 1, they are arranged side by side with the hadrom (H) on the outside and facing each other with the leptomal portion (L). The veinlets originating from one of them enter both the minor and major wings on the same side, showing that the minor and major wings on one side represent in fact the lateral halves of a blade, whose midrib is probably represented by one of the vascular bundles. When the malformation advances further, the four wing-like processes pass into the four-lobed blade (Figs. 19, 20) and the latter in turn into the dichotomous blade (Figs. 21, 22).

Taking the relative position of the leptomal and hadromal portion in the vascular bundle of the blade and the position of the caeomasori on the blade into consideration, it may be concluded that the inner wall of the anther-lobe corresponds to the upper surface of the leaf, if we admit, according to the view above set forth, that the anther-lobe is produced by the bulging out of a leaf at its apical portion.

Cases of bilaminar malformation have been reported in the ordinary leaf. MASTERS ('69, p. 446) gives an instance in the orange tree, in which some leaves presented the appearance of having been formed by the fusion of two leaves back to back and with the vascular bundles in the common midrib arranged in a circular manner and not in a horse shoe-like arrangement. BUCHENAU ('88) describes a four-winged leaf in tobacco, which is formed, according to his explanation, by an excres-

cence of the blade. A. ERNST ('99) gives two instances of laminar enation. In *Anthurium* he found a boat-like adventitious lamina on the under surface, arising midway between the primary nerves with its inner surface corresponding in its structure with the upper surface of the primary lamina. He ascribed this malformation to the bulging out of the internal tissue towards the under surface of the leaf. In a mango-leaf he found also a secondary leaf of a little boat-like shape on the under or upper surface of the primary leaf, adhering with its keel to the midrib of the latter. According to his statements, the secondary leaf has a median nerve which is proved evidently to be a piece of the outermost fibro-vascular bundles of the original costa. If I understand him aright, the secondary leaf, differently from what we saw in *Prunus*, seems to have its veinlets originating from the median nerve, while the original costa ramifies in the primary blade. We see in these cases that the mode of formation of the accessory leaf is different from that of the bilaminar staminal leaf, and so we can not consider the given malformation of the anther as an abnormality without regularity as is usually assumed in similar malformations of the ordinary leaf.

In the present case the malformation of the anther goes on in different ways according as it undergoes phyllody or petalody. In the so-called double flower (*flores pleni*), the petalody of the stamen is simpler than the phyllomorphic change, the anther-lobe taking no essential part in forming petaloidal stamens. As the first step of the change, there appears a small ligule projecting from the apex of an ordinary anther, which appears to correspond to the expanded connective. When the change proceeds further, the ligule becomes larger and larger, till it assumes a petaloidal appearance as well as form, while the anther-lobes are borne on its upper surface in an abortive form. The filament becomes more and more laminar, and in a completely petaloidal stamen it constitutes the lower part of the petal (Figs. 37-40). Essentially similar changes are found in the petalody of the stamen of *Camellia japonica* (CELAKOVSKY, '78; compare his Figs. 14-35).

Malformation of the Ovule.

In a full grown and completely phyllomorphic carpellary leaf, we can hardly point out the portion corresponding to the ovule. But in some chloranthic flowers, there are found at young stages some carpels in various degrees of malformation, which appear to me to throw light on the morphological value of the ovule.

In general, the degree of change of the ovule seems to correspond to that of the pistil to which it belongs. Examples of the least deformed pistils are shown in Figs. 2, 44, and 45. Compared with an ordinary pistil (Fig. 43), the essential external difference lies in the hypertrophied, fusiform ovary; the malformation is little advanced, and the outer surface is covered with the same hairs that characterise the normal pistil. Opening such a pistil longitudinally, we find certain changes inside the ovary. A pair of much hypertrophied ovules are developed on the ligulate process appearing along the ventral suture line (Fig. 45). Below them may be seen a pair of serrated processes. The whole condition probably represents the first step of phyllodic metamorphosis.

The next stage of malformation is found in a similarly deformed pistil (Fig. 46). There is nothing important in its outer appearance that distinguishes it from the foregoing, but the ovules, instead of being solid cell masses, are here represented each by a slightly parted lobe, which however looks, when rolled up, like a solid ovule as before. In the centre of such a lobe is found a small papilla (Fig. 47). A full grown ovule in a similar state of malformation is often found in a completely phyllomorphic carpel (Fig. 51), and in such cases the division of the ovule into the outer laminar part and the median papilla may be most distinctly seen. On tracing back the steps through which the ovule has passed in this process of malformation, we find that the papilla corresponds to the nucellus and the laminar part to the primine (integument), and that the laminar part has a tendency to hypertrophy, while the papilla shows the opposite tendency.

The most advanced malformation so far observed in a closed carpel is shown in Fig. 48. The lamellar process developed along the suture is more prominent than before, and the position of the ovule, or the portion of the process corresponding to it, is not easily discernible. We are inclined to the view that the lamellar process or processes developed both above and below the ovules, as shown in the preceding figures, approximately represent the margin of the leaf, while the laminar ovules have themselves undergone similar changes, so as to bring ultimately the differences between the two parts to nil.

A more advanced stage of the malformation of the pistil is shown in Fig. 49. Though the general form does not differ from those of the preceding examples (Figs. 44-48), the ventral suture shows a slit, and the margin of the carpel becomes visible externally. Opening this carpel we find a pair of bifurcated lobes at the place corresponding to that of the ovule (Fig. 50). As such a carpel is found in completely chloranthic flowers, I am sure that it can develop later into a completely phyllomorphic leaf, such as is shown in Fig. 52 or Fig. 53, where the marginal lobes that have replaced the ovules can still be pointed out. It may be noted that the bifurcation of the ovular lobes is already apparent at a very early stage of development and remains so until a somewhat advanced stage (compare Fig. 50 and 53), and the single lobe found in place of the ovule (Figs. 52, 54) indicates probably the form more advanced in phyllody than the bifurcated one.

In the most completely phyllomorphic carpel the ovular lobe becomes less conspicuous and finally indistinguishable from the serration of the margin. This is so not only in the adult leaf, but already in a young stage, as shown in Figs. 3, 4.

The series of transitional forms between the normal and malformed ovule above described, in which the ovule appears first as a lobe on the carpellary leaf and, becoming generally less prominent, finally forms an integral part of the carpel, will justify us in concluding that the ovule in fact undergoes the phyllomorphic change during the metamorphosis of the carpel.

Keeping this fact in mind we will inquire shortly into the morphological nature of the ovule and its covering. Indeed this is a difficult problem, and various conflicting views are still prevalent among eminent botanists. According to WORSDELL'S ('04) review of this subject, the various published opinions may be classified into three classes, viz. (1) the axial, (2) the foliar, and (3) the *sui generis* theory. The arguments advanced in support of these theories are based essentially on the developmental, comparative, anatomical, or teratological studies of the ovule, or on a combination of them, and the different opinions of the authors seem to depend on the comparative weight which they attach to different data. I have no intention of generalising my observations on *Prunus* into a theory, but if we once admit the value of teratological evidences in this case, as CELAKOVSKY ('84) so earnestly contends for, there is no doubt that the facts presented above agree best with the foliar theory¹. For, we have obtained several transitional forms from the entire ovule to the marginal lobe of the carpel and even from the lobe to the serrated condition. Hence, if the malformation in this direction be taken as of equal weight as the phyllody of the pistil, in determining its morphological nature, we are led to look upon the ovule as a process or lobe developed from the margin of the carpellary leaf, and not a new growth or bud. The lobe derived from the malformed ovule corresponds to its integument, while the nucellus, the reproductive organ proper, is replaced by a small papilla which undergoes complete atrophy when the malformation advances further. The nucellular portion leaves no corresponding portion on the phyllomorphic carpel; it may therefore perhaps be a new growth.

Etiological Considerations on Chloranthy.

Having given a detailed account of the possible changes of the flower caused by *Caeoma Makinoi*, we shall now proceed to consider, from the

1. A historical sketch of the various views on the morphological nature of the ovule is given by WORSDELL ('04) in a concise but very comprehensive form.

the etiological point of view, the relation between the development of the fungus and the chloranthic change. Respecting the cause of this change in general, several views have been expressed by different authors. Some (FRANK, '81, p. 436; GOEBEL, '84, p. 124; SORAUER, '09) maintain the view that plants are inclined to undergo chloranthic or similar pathological malformation when subjected to excessive humidity or nutrition. This view agrees to a certain extent with the phenomenon that the over-nourishment of the vegetative organs generally tends to inhibit the formation of the reproductive organs, leaf-buds, for example, replacing flower-buds. MASTERS ('69, p. 280) has already remarked, "It might at first be supposed that the same causes that bring about the complete substitution of leaf-buds for flower-buds would operate also in the partial substitution of leaves for other parts of the flower, but it will be seen that the inducing cause, whether similar or not in the two cases respectively, acts at different times; in the one case, it is not brought into play until the rudiments of the flower are already formed, whereas in the other the influence is exerted prior to the formation of the flower. So that while the formation of leaf-buds in place of flower-buds may be and generally is due to an excess of nutrition, inducing overactivity of the vegetative organs, the production of phyllomorphic or chloranthic flowers may be owing rather to a perversion of development arising from injury or from some debilitating agency. The discrepancies in the assigned causes for the conditions above mentioned may, therefore, in great measure, be attributed to the different periods at which the causes in question operate." In studying the tuberous change of flowers in a *Nymphaea*, BARBER ('89, p. 113) holds a similar view as its cause. Thus, according to the last mentioned two authors, the important factor for chloranthic change is the period at which a certain cause operates, and, therefore, the cause which perverts the formation of the flower may equally give rise to chloranthic flowers, if it operated later.

In a great majority of cases, chloranthy is caused by a certain physiological disturbance prevailing in the whole plant body, whereas the time or duration of the action is not clearly conceivable. The question

is, however, quite different when the initial change is due to parasites, whether insects¹ or fungi. In these cases there is clearly a time and spatial relation between cause and effect. The study of the close relation between the parasite and the induced change seems, however, to have been neglected. PEYRITCH ('83) first made an experimental study of the chloranthly caused by the attack of an insect. After numerous experiments he came to the results that the degree of the change produced is proportional to the number of the insect concerned and the period of infection of the flower-bud. Even with this valuable investigation the extent of the stimuli of the insect is not yet thoroughly clear. As to the intimate cause of the general malformation of flowers, the hypothesis of SACHS ('93, p. 236) may be mentioned that the formative substances of floral organs, which are produced by the leaves and determine definitely each floral organ, are in normal development distributed with mathematical accuracy to still embryonal organs; but "Abnormitäten", he says, "dadurch hervorgerufen sein können, dass in der mikroskopische kleinen jungen Blütenknospe einige organbildender Substanz einen unrichtigen Weg genommen oder zu spät oder zu früh eingewandert sind u.s. W." GOEBEL ('98, p. 174) seems to hold a similar view concerning certain kinds of malformation.

The first question to be considered in the chloranthly before us with regard to the relation of the fungus and the malformation is that of the distance to which the action of the fungus may extend in the surrounding tissue. It is known that in the branch of the witches' broom, usually caused by parasitic fungi, the leaf-bud is substituted for the flower-bud. In this case, the mycelia traverse the whole diseased branch, so that any abnormal development seems to be correlated with the whole organisation of the broom. An important point of difference in the present case is that the fungus development is generally limited to the bud, and the abnormal development of the latter is to be assigned to the

1. Numerous instances of the phyllody of some floral organs caused by animals are enumerated by V. SCHLECHTENDAL in "Die Gallenbildungen 1891."

mycelium found in its tissue. Even in a floral organ attacked by the fungus the phyllomorphic change is sharply limited to the region occupied by the mycelium; for instance, a portion of the affected corolla remains quite healthy, if it is not invaded by the mycelium (Fig. 2). It appears highly probable that the infection of the floral organ by the fungus is by no means the necessary cause of phyllody; the malformation is observed only in those parts which are traversed by the mycelium.

As we have already stated, the degree of malformation is correlated with the mode of the fungus development. Although it appears that the intercellular mycelium would determine, first of all, an alteration of the course of development of the floral organ or its portion leading to phyllody, the subsequent development of the latter, attended by corresponding malformations, must be associated with the manner of the fungus development. Were the fungus under condition incapable to complete its development, the phyllomorphic change is less remarkable. On the other hand, typical malformation always accompanies the formation of the caeomasori. This fact gives us the impression that the stimulus of the fungus is stronger when the caeomasori appear.

The most important point to be considered from the etiological point of view is the period of infection of the fungus. The bud may be infected rarely by the perennial mycelium and, in the majority of cases, by the spore. Internal infection by the mycelium takes place, when a bud occurs within the extent of the intercellular mycelium which has spread up and down, chiefly through the cortex, from the node where an affected bud was developed the preceding year. In the case of external infection it is still obscure what kind of spores effects it, since the life-history of the fungus is not clearly known; so that the period of infection can not be determined accurately. On the other hand, I think that some ideas on the subject may be formed from a consideration of the behaviour of the perennial mycelium.

Among the flower-buds appearing successively at different distances from the node, the bud of which was affected the preceding year, and from which the perennial mycelium has spread out through the cortex

of the stem, chloranthic malformation is seen only in a certain number of them lying near the node. Of these, again, the degree of malformation decreases successively from the bud nearest to the node to the one lying farther from it. Keeping in mind that the diseased bud, whether weakly or strongly affected, must be, as already stated, always invaded by the fungal mycelium, and considering that the arrival of the mycelium in each bud occurs at different times, it can be conceived that the difference of malformation they exhibit is due to the different periods of infection. This view is supported by the peculiarities of the diseased branches shown in Text-fig. 2.



Text-fig. 2.

The branch A was infected by the fungus the preceding year at the node 1, and the flower (*f*) and shoot (*s*) derived from it were affected. Above and below this node the stem shows a slight swelling, and a longitudinal fissure (*r*) is produced on the cortex, extending to the node 2. The cortex is here traversed by the intercellular mycelium which sends

out, at intervals, cystolith-like haustoria in the surrounding cells (KUSANO, '06). The mycelium, proceeding further up the branch from the node 2, reaches a higher flower-bud on it. The bud develops a flower in partial chloranthy: healthy whitish portions are seen on the petals, the stamens carry normal anthers, and the pistil has a closed ovary. The flower on the branch from the node 3 upwards is quite healthy, as it lies beyond the reach of the mycelium.

Similar facts may be seen more clearly in the branch B. It was infected the preceding year at *a*. In the same year a healthy flower was developed at *b* and also the normal shoots, *x* and *y*. The mycelium entered the branch at *a* and has thence spread up and down through the cortex and caused an abnormal thickening of the branch, more markedly at *a* and *b*. The fissure in the cortex extends in the shoot *x* beyond the bud *g*, and in the shoot *y* below the bud *d*. The buds of these shoots are affected in different degrees. The bud *c*, which appears, from its position on the shoot, to develop into a flower, shows the greatest distortion, and gives rise to a diseased shoot. The symptom of its affection was visible already in January by its unusual enlargement and by having succulent loosely over-lapping bud-scales. It is certain in this case that the mycelium has invaded the bud when the rudiments of the floral organs, which would be formed under normal conditions, were still undifferentiated, and thus inhibited its development into a flower. The bud *d* on the next node develops a completely chloranthic flower: in the embryonal form the stamens and pistil are entirely leafy. The change is incomplete in the bud *e*, developing an imperfectly chloranthic flower. Lastly the bud *f*, lying beyond the reach of the mycelium, grows into a quite healthy flower. Something similar is observed in the shoot *x*: a completely chloranthic (*g*), incompletely chloranthic (*h*), and normal flower (*i*) are arranged successively in the lower and the upper portion of the shoot.

It is very evident that the extension of the mycelium from the infected point is gradual, and the more proximal buds are infected earlier than the more distal ones. The incompletely chloranthic malformation

of the flower on the distal portion is, therefore, due to a later infection by the mycelium, the infection having taken place after the rudiment of each floral organ had already been formed. It is also quite natural that, if the mycelium invade the bud at the time when its embryonal processes are not yet differentiated into floral organs, a diseased shoot (*c*) would appear from it, while a slightly later infection makes the foliage leaves derived arrange in whorls as the normal floral organs.

I may here make an attempt to determine the exact time of the year when the mycelial infection of the bud is efficient enough to cause chloranthly. According to MASTERS' view and from the facts we have just referred to, it may be inferred that the question would be solved, if the period at which the embryonal processes in the bud are definitely differentiated into floral organs be accurately ascertained. So far as has been observed in Tokyo, this period is attained before the vegetative season is over; in the autumn (October) the rudiment of the floral organs are already formed distinctly and arranged in whorls. Hence, if the mycelium should begin to exert its malforming action for the first time at or after this period, the infected flower can not show anything else than incomplete chloranthly, developing, for instance, such staminal leaves as are shown in Figs. 23-26.

Again, in the early part of the vegetative season (June or the end of May), while the new shoot is yet at the growing stage (Fig. 5), the rudiment of the winter bud is not visible, so that, if the mycelium should at this period reach the axil of a leaf, the bud to be developed there subsequently would be primarily affected, and produce a shoot instead of a flower. From these observations it may be concluded that the period of infection by the mycelium, necessary to cause typical chloranthly, is between the late summer and early autumn, most probably in September or August.

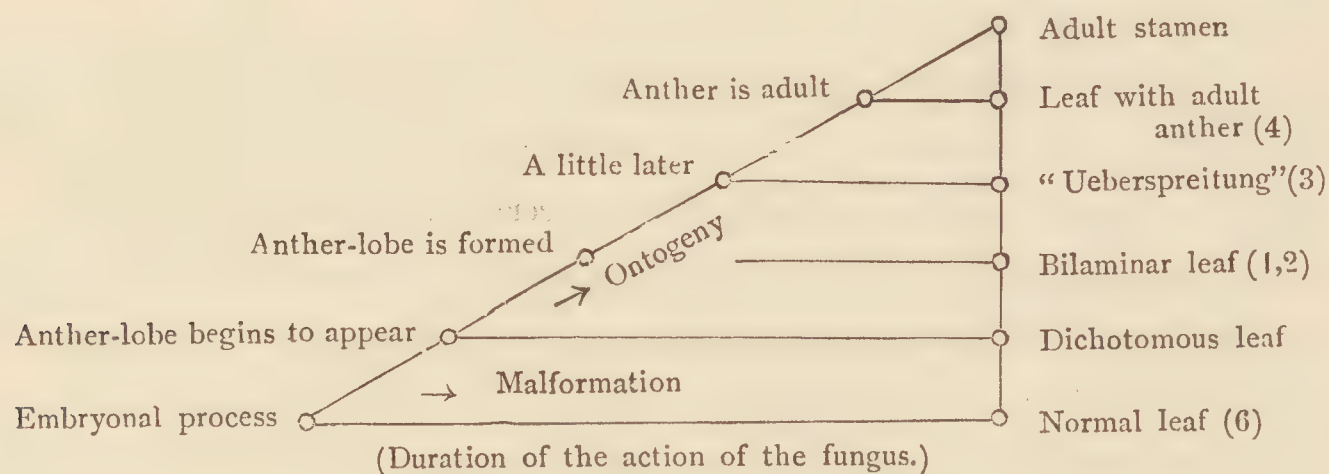
Turning again to the branch B in Text-fig. 2 with this consideration in mind, we may make the following statement about the development of the mycelium through the mother-branch of the buds as follows:

While the affected bud at *a* is growing into either a chloranthic flower or a diseased shoot and its sister-buds are being developed into the normal shoots *x* and *y*, the mycelium at *a* begins to spread with great rapidity. It is in June or July that the mycelium invades the basal portion of these shoots, where the rudiment of the bud is not yet formed. The bud *c* is perhaps formed after the arrival of the mycelium. The arrival of the mycelium at the nodes *d* and *g* takes place still later, perhaps in August, when the buds have already developed far enough to form the floral organs. Further, it may be assumed that the buds at *e* and *h* have advanced in normal development so far, previous to infection, that the subsequent action of the mycelium is no longer efficient to produce complete phyllody.

In the chloranthy due to external infection by the spore, we can find also several forms of malformation. Leaving the question as to what kind of spores the infection is due to for further investigation, I may at present propose the view that the period of infection, or strictly speaking, of the beginning of the malforming action, must be the same as in the case of the mycelium. The spore attaches itself to the axil of the growing shoot and germinates there in June or still later. Some spore may germinate previous to the formation of the bud-rudiment, while some may germinate after the bud has formed the embryonal floral organs. According to the earlier or later germination of the spore, we will obtain from the affected bud a shoot instead of a flower, or a completely or incompletely chloranthic flower.

The conception we have thus developed as to the relation between the degree of malformation and the period at which the perverting action of the fungus begins may also be drawn from a chloranthic flower itself, by considering the different amount of change exhibited by the phyllo-morphic stamens mentioned before. The staminal leaf with the upper half of the stamen unchanged may be the result of an infection which took place at the moment when the proper form of the stamen was nearly completed, but a meristematic portion was still left at its base, the mycelium having attacked this meristematic portion. A slightly earlier

infection would result in the formation of a structure with four wing-like processes in place of the anther. On the other hand, a complete phyllody will result when the fungus begins to act upon the stamen while it is still an undifferentiated process. On the whole, we agree with GOEBEL (98, p. 155) who says, "Es kommt also bei der Umwandlung von Staubblattanlagen in Betracht erstens die Entwicklungsstufe, auf welcher die Staubblattanlage steht zur Zeit, wo sie den Antrieb zur Umwandlung—wenn dieser Ausdruck gestattet ist—erhält, und auf die Grösse dieses Antriebs." Hence the several forms of the malformed stamens mentioned in the foregoing are the results of infection at different stages when the normal development of the stamen became affected. This relation may be represented in the following diagram¹:



From this arrangement we can see that the phyllody of the stamen, at least in the present case, is not a capricious monstrosity, but that the several forms of the malformation appear to be exaggerated representations of the forms which the stamen assumes at different stages during its ontogenetic development.

The evidence that the degree of malformation is associated with the stage of development, at which the perverting action begins, is also afforded by the different organs of the same chloranthic flower. As the lower member of the floral organs precedes in development the upper one, it happens that, when the upper members is in the meristematic condition

1. Arabic numerals in the diagram give the types of phyllomorphic stamens described before.

at the time of infection by the fungus, the lower one has already passed on to a more advanced stage, and offers a greater resistance to the malforming action, the result being that lower one suffers a lesser degree of malformation. So in any chloranthic flower the pistil is more advanced in phyllody than the stamens, or the stamens than the petals and sepals (Fig. 3). Again in a lately infected flower (Fig. 2), we see that the pistil shows the greatest malformation, while the stamens or petals present only a slight deformation in their basal portions. Further, among the stamens, those of the inner whorls show a greater degree of malformation than those of the outer; for instance, bilaminar or dichotomous leaves inside the ordinal simple leaves, the leaves with phyllomorphic anther-lobes inside those with the anther and the upper portion of the filament unchanged, etc.

Lastly we will consider in connection with our subject the less affection of the bud-scales. That the outer scales, in most diseased buds, are free from the attack of the fungus is certainly due to the completion of its development before the fungal infection. Though the inner scale is usually affected, even at an early time, its subsequent development is completed prior to the beginning of the next vegetative period, in other words, before the vigorous development of the infected mycelium begins, and hence a slight malformation results. The infection of the bud occurs too late to induce an abnormal development of the scale, and if the infection takes place at a sufficiently early period so as to induce on the scale a notable malformation, the bud would never become a chloranthic flower, but would develop into an affected shoot, as in the case of the bud *c* in Text-fig. 2.

On the basis of these facts I may conclude that the fungus shows in its chloranthic action a certain parallelism with some insects. According to PEYRITCH ('83), the degree of the changes, similar to those dealt with in this paper, of *Arabis* flower when attacked by an *Aphis* depends upon the stage of its development, at which the insect becomes parasitic, and also upon the number of the insect found on one flower. His conclusion is based on the observation of the changes which the flower

passes through, when various numbers of the insect are placed on it at different stages of its development. Although I was not able to carry out a similar experiment with the fungus, yet the careful observations of numerous diseased flowers, especially such as are shown in Text-fig. 2, are, in my opinion, sufficient to justify a conclusion which is in strict accordance with PEYRITSCH's view respecting the degree of malformation in relation to the developmental stage of the affected flower. As to the intensity of the stimulus, he placed the principal weight upon the number of the insect, but I have to compare with it the condition of the development of the fungus: the same condition that inhibits the formation of caeomasori produces a slighter degree of malformations.

As PEYRITSCH has pointed out, a too late infection does not produce chloranthly. The same may be true for the fungus parasite, if we assume that, in the case of the insect, the more developed flowers do not respond to the stimulus of the insect, while in the case of the fungus, the flower at the corresponding stage does not allow the fungus to spread further through its tissue and consequently to exert a stronger stimulus.

In our case chloranthic change goes hand in hand with an extraordinary hypertrophy. The flower must be overnourished. Whether there is causal relation between the two phenomena is at present not certain. It may, however, be that, under these conditions, the fungus is able to develop more vigorously and to exert a stronger stimulus.

General Remarks.

The chloranthly of *Prunus Mume* is due to the action of the fungus parasitic in the flower-bud. The protoplasm of the undifferentiated organ reacts to the stimulus exerted by the mycelium and the course of development is modified in the way mentioned. The stimulus of the mycelium for causing chloranthly is very limited in extent. The development of chlorophyll, attended by other external and internal modifications of the floral organs, extends *pari passu* with the extension of the mycelium. A strong malformation is correlated with a vigorous development of the

fungus, so that, when the conditions are such as to inhibit the full development of the latter, the infected portion of the flower undergoes less malformation.

Complete chloranthy does not result unless the necessary action of the fungus operates upon the bud at a certain stage of its development. Its operation at earlier stages develops the bud into a shoot, while at later stages it produces incomplete or no chloranthy.

The eminent morphologist VELENOVSKY ('05, p. 20) has distinguished the abnormalities occurring in plants into a few categories¹. According to him, the present instance should be included in the pathogenetic abnormality, since he defined it as being caused by insects, fungi, wound, or injurious chemical agents. However, he admits that "alle vergrünnten Blüten" (p. 24) belong to the category of the morphological abnormality, on which he places considerable importance in developing his views on the morphological nature of plant organs. It seems, therefore, that the malformation of anthers and ovules presented by our chloranthic flower may be a welcome addition to our knowledge of the morphological nature of these organs for those who, like VELENOVSKY, attach much weight to abnormalities in the question at issue. As for myself I look upon abnormalities discussed above as atavistic phenomena, and am of the opinion that chloranthic flowers throw some light on the morphological nature of floral organs in general.

Reviewing now, as a whole, phenomena of the chloranthy and the associated changes caused by the fungus, the relation between the time of infection and the degree of the ensuing malformation will become very apparent. This may be easily seen from the following series of diseased buds and their derivatives thus far obtained by me:

- a. Bud looks like a normal leaf-bud, outer bud-scales attacked by the fungus; develops into a shoot.
- b. Bud swollen just like a flower-bud, outer scales healthy; develops into a shoot.

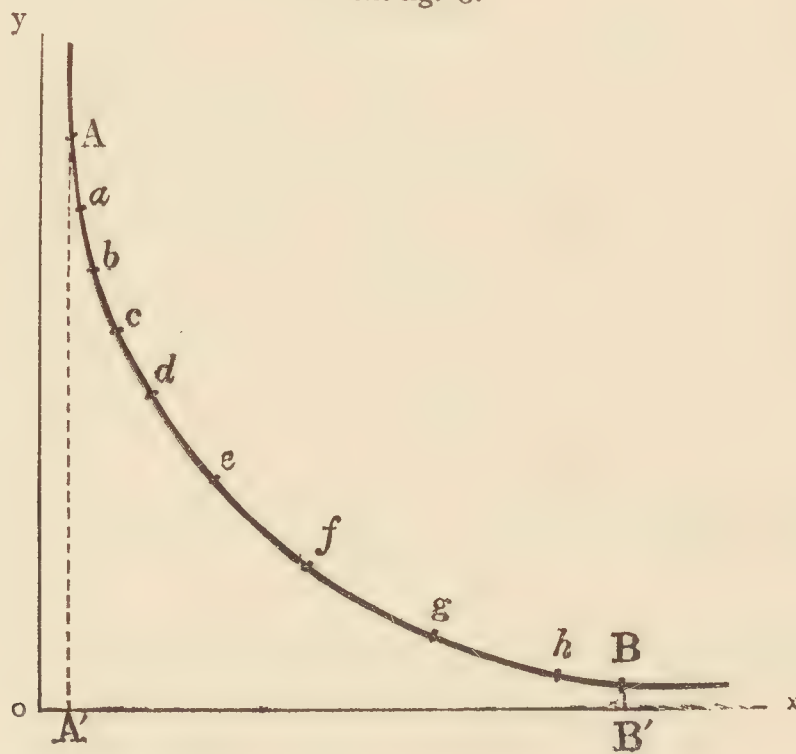
1. "Pathogene, Extrem-, durch Wurderung entstandene und morphologische Abnormalitäten."

- c.* Bud swollen just like a flower-bud, inner scales look like sepals; develops into a shoot.
- d.* Chloranthic flower: axillary and median proliferation.
- e.* Chloranthic flower: stamen into dichotomous or simple leaf.
- f.* Chloranthic flower: stamen into bilaminar or double leaf.
- g.* Chloranthic flower: stamen into a leaf whose upper half is unchanged.
- h.* Partially affected flower: slight change in the basal portion of each floral organ.

In *a*, which otherwise would have developed into a flower-bud, the infection takes place at the earliest stage, and in *h* at the latest stage, of the development of the bud. A still later infection than in *h* is not effectual, since the floral organs have become resistant to the attack of the fungus.

The several degrees of malformation mentioned above lead me to say that the change is greater if the time of infection is earlier, and smaller if it is later. Denoting now the time of infection by x , an earlier infection being expressed by a small value and a later infection by a large value of x , and the amount of change by y , we may say that y is inversely proportional to x , that is, $y = \frac{k}{x}$ or $xy = k$, where k is a coefficient.

Text-fig. 3.



It is seen from this equation that the change caused by the fungus is expressed by an hyperbola whose asymptotes are $x=0$ and $y=0$, assuming k to be a constant¹. So that, in the supposed curve (Text-fig. 3) greater change lies near the point A and less change near B . That a too early and too late infection brings always results in a constant change, the former giving a shoot and the latter a normal flower, is evident when we note that the decrease or increase of x beyond a certain limit (A' and B') gives nearly a constant value of y . This assumption is not inconsistent with the nature of hyperbolic curve, since the curve cuts the asymptotes at an infinite distance; in other words, the extremities of the curve run parallel to the asymptotes.

1. The coefficient k may be variable according to the conditions of the development of the host and fungus, or of the environment. We assume here all these conditions to be constant.

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1. The asterisk marks those to which I have not been able to gain access.

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EXPLANATION OF PLATES.

All figures were drawn from fresh materials and, except Figs, 45-48, 50, are reproduced in natural size.

Plate XVII.

Fig. 1. Young diseased shoot with some inner bud-scales carrying spermogonia.

Fig. 2. Partially affected flower, not essentially deformed.

Fig. 3. Young chloranthic flower having partly phyllomorphic stamens; three normal flowers already pollinated, and three normal leaf-buds.

Fig. 4. Young chloranthic flower having completely phyllomorphic stamens and affected bud-scales.

Fig. 5. Chloranthic flower in full growth, young fruit, and normal shoot, all developed on a shoot of the preceding year. The yellowish-orange caeomasori have densely covered the upper surface of all the blades of the flower.

Plate XVIII.

Fig. 6. Much hypertrophied stalk of a chloranthic flower with diseased bud-scales at its base.

Fig. 7. Outer healthy scale of a diseased leaf-bud.

Figs. 8, 9. Partially affected inner scales of the same, a slight thickening taking place at the portion occupied by the spermogonia.

Fig. 10. Slightly phyllomorphic sepal, not producing caeomasori but only spermogonia.

Fig. 11. Slightly phyllomorphic petal producing caeomasori only in one portion.

Fig. 12. Petal with the apical portion free from caeomasori and less deformed.

Fig. 13. Much enlarged petal undergoing different deformation in portions occupied and not occupied by the caeomasori.

Figs. 14-36. Various phyllomorphic stamens.

Fig. 14. Anther-lobes expanded into four wing-like processes, and the basal portion of the filament undergoes a somewhat laminar deformation.

Fig. 15. The anther is replaced by double blades which are connected with the main blade derived from the filament by a narrow neck. *a*, under side; *b*, upper side.

Fig. 16. The entire stamen is represented by two blades, coherent, as it were, along their midribs.

Figs. 17, 18. The apex of the double blades bears a papilla-like process.

Figs. 19, 20. Lower portion of the filament becomes the main blade and an accessory blade derived from the anther-lobes rests on it with a short stalk.

Figs. 21, 22. The anther is replaced by a dichotomous leaflet, while the basal portion of the filament is mainly affected by phyllomorphic change.

Figs. 23-26. Lower half of the filament partakes of phyllody, while the upper half including the anther shows no essential change.

Figs. 27-29. Phyllomorphic change takes place independently in the upper and lower halves of the stamen.

Figs. 30, 31. Three-lobed staminal leaf.

Fig. 32. Peltate staminal leaf.

Fig. 33. Spatulate staminal leaf.

Fig. 34. Oblanceolate leaves representing entire stamens, coherent with each other along their petioles.

Figs. 35, 36. Dichotomous leaf representing an entire stamen.

Figs. 37-40. Petaloidal stamens in various degrees of change.

Figs. 41, 42. Shoot proliferated from the axil of a stamen.

Fig. 43. Healthy pistil shortly after fertilisation.

Fig. 44. Pistil of the partially affected flower shown in Fig. 2, with spermogonia on its surface.

Fig. 45. The same dissected, showing two ovules in the first stage of malformation. Above and below the ovules are seen serrated lobes along the suture line. $\times 2$.

Fig. 46. Similar pistil, with the integument of the ovule unfolded into a slightly bifurcated lobe. $\times 2$.

Fig. 47. Ovary of an affected pistil, opened longitudinally along the suture line. In the place of the nucellus appears a papilla-like process, resting on the laminar integument. *a*, outer view of the marginal portion of the carpel folded in the ovary; *b*, inner view of the same. $\times 2$.

Fig. 48. Similar ovary opened. The ovules become marginal lobes. $\times 2$.

Fig. 49. Young phyllomorphic carpel.

Fig. 50. The same with the margins spreading out laterally. Serrated lobes in place of the ovules are the largest. $\times 2\frac{1}{2}$.

Fig. 51. Full grown phyllomorphic carpel. Ovules are not yet completely phyllomorphic and the nucelli are shown as papillae on an incompletely laminar integument. *a*, *b*, upper view of the ovules on the left and right side of the carpel respectively.

Fig. 52. Full grown phyllomorphic carpel provided with stipules and lobed margin. The largest lobe on either side occupies the position of the ovule.

Fig. 53. Gigantic phyllomorphic carpel from a flower, in which most of the floral organs have atrophied. A large irregularly parted lobe represents the malformed ovule.

Fig. 54. Trilobate carpellary leaf. Lateral lobe represents the malformed ovule.

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Vol. II. No. 6, PUBLISHED APRIL 23th, 1911.

Price in Tokyo Yen 0.39

THIS JOURNAL IS ON SALE AT

Z. P. Maruya & Co., L'td.

Tori Sancho, Nihonbashi, Tokyo.

Geiser & Gilbert.

Kaji-cho 23, Tokyo; Main Street 77, Yokohama.

Josephinenstr. 6. Leipzig.—R., Germany.

明治四十四年四月二十三日印刷
明治四十四年四月二十五日發行

編纂兼發行者 東京帝國大學

印刷者

東京市麹町區內幸町一丁目五番地

松浦彦一

印刷所

東京市麹町區內幸町一丁目五番地

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NOTICE.

VOL. I.

- No. 1, published June 16th, 1909.
- No. 2, published June 29th, 1909.
- No. 3, published March 28th, 1911.
- No. 4, in press.

VOL. II.

- No. 1, published December 8th, 1909.
- No. 2, published February 12th, 1910.
- No. 3, published April 30th, 1910.
- No. 4, published July 18th, 1910.
- No. 5, published October 31st, 1910.
- No. 6, published April 25th, 1911.
- No. 7, in press.

VOL. III.

- No. 1, published August 30th, 1910.
 - No. 2, in press.
-

COI

13,851

Vol. II. No. 7.

東京帝國大學
農科大學紀要

第二卷第七號

JOURNAL

(FORMERLY BULLETIN)

OF THE

COLLEGE OF AGRICULTURE,

IMPERIAL UNIVERSITY OF TOKYO.

TOKYO.

Published by the University.

A DECEMBER 26TH, MEIJI XLIV (1911).

PUBLISHING COMMITTEE.

PROF. Y. KOZAI, *Nōgaku-hakushi*, Director of the College (*ex officio*).

PROF. C. ISHIKAWA, *Ph. D.*, *Rigaku-hakushi*.

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All communications relating to this Journal should be addressed to the
Director of the College of Agriculture.

Prehistoric Fishing in Japan.

BY

Kamakichi Kishinouye.

College of Agriculture, Komaba, Tokyo.

With Plates XIX—XXIX and 10 Text-Figures.

The people of prehistoric ages have obtained the necessities of their daily life chiefly by hunting and fishing, and these two occupations were at the same time the principal pastimes for these primitive people. Consequently in those times hunting and fishing were comparatively well developed, and fishing was especially important to insular inhabitants. In prehistoric times the Japanese Islands were inhabited by a piscatorial people belonging to a race quite different from our ancestors.

According to the recent progress of Anthropology in Japan, shell-mounds have often been dug out and well explored, and fishing implements, fish remains and other objects relating to fish and fishing were collected in pretty large number. Collections of these objects have been made by members of the Anthropological Institute of our University, Prince NIJO, Messers KICHIBEI NAKAJIMA, SOICHIRO MORI, ICHIHEI ONO, SHINZO SUZUKI, TAMEJI TAKASHIMA, SUIN YEMI, etc. besides the author. These gentlemen have assisted me in the preparation of this paper by loaning or granting specimens and by giving much valuable information, and I wish to express here my sincere thanks for their kindness.

All articles found in these collections belong to the so-called Neolithic Epoch and most of them belong to sea-fisheries. They were chiefly obtained from shell-mounds associated with many stone-implements, crude

potteries, hard parts of molluscs, fishes, turtles, birds, many marine and land mammals, etc. Fishing implements frequently discovered are arrow-heads, spear-heads, harpoon-heads, hooks and sinkers. A few canoes of primitive types were also recovered from alluvial plains.

Remains of shellfish, fish and other aquatic animals are very plentiful and most of them belong to the marine fauna. We do not know whether the prehistoric people have utilized algae or not. Fragmentary exo-skeletons of sea-urchins and crustaceans; a great many kinds of molluscan shells; cuttles of cuttlefish; scales, teeth, spines, bones, otoliths, etc., of different fishes; bones of turtles; and teeth and bones of seals, whales and porpoises are often found. Only a few forms of purely fresh-water animals, such as *Corbicula*, *Anodonta*, carp, etc. have hitherto been found. We must not think, however, that all animals, remains of which are found in shell-mounds, were captured and utilized by the prehistoric people who made the mounds. Some animals or their skeletons may have been stranded and then utilized, and some kinds or their skeletons were probably imported from other regions.

Remains of fish and fishing implements investigated by me were mostly collected from the districts around Tokyo and the regions north from the capital. From these materials we find, that prehistoric fishing in our country was wonderfully well developed.

Shell-Mounds and Fishing Villages.

Shell-mounds are the principal sources of materials from which we derive our knowledge of prehistoric fishing. These shell-mounds are found here and there along the coast and they are especially abundant in the Kwantō and the north-eastern districts of Hondo. In these districts several shell-mounds are found, crowded often for a distance of a few miles. We find them even on small isolated isles, pretty far off in the sea, for example on Oshima near the mouth of Tokyo Bay. Shell-mounds differ greatly in magnitude. Some of them are enormously large, covering an area of several square kilometres, with a thickness of several metres. They are

generally found several metres higher than the present sea-level, thus in flat countries they are several kilometres inland from the present sea-shore. In a few cases the greater part of the remains in mounds consists of fish-bones, molluscan shells being comparatively rare, and in a site at Misaki, Kanagawa-ken, Mr. S. YAGI found fish-bones only.

The situation of fishing villages in prehistoric times may be imagined from that of shell-mounds and we find that the site selected for villages was more or less convenient for anchorage or near sandy shoals, where shell-fish grow in large quantities. As different shell-mounds contain more or less different kinds of aquatic animals and fishing implements, it seems that the inhabitants of different villages had different tastes and different talents in fishing, as it is also the case at present. No doubt there were some means of communication among these villages, as fishing implements and other utensils were more or less alike to each other in villages situated near together. Villages on small isles, however, were generally rather isolated.

Missile Implements.

The missile implements are simple, convenient, and consequently most popular among primitive fishermen, as it is told that man hunted fish before he caught them. The implements of this kind are well developed and are dexterously used in the Stone Age; thus we find a great many varieties in our prehistoric fishing in the form of arrows, darts, spears and harpoons.

ARROW-HEADS AND DART-HEADS.

Stone arrow-heads are found in shell-mounds as well as in other places where no fish-remains occur, and there are a great many kinds in form, size and material. There is no doubt that at least a portion of them was used for the capture of aquatic animals. Really we find that the apical portion of some fishing implements were made of stone, so that the apical portion of harpoons, etc., made of stone is included in what we simply

call stone arrow-heads. Stone arrow-heads are about 15—50 mm. in length, triangular, rhomboidal, lanceolate, etc., in form and made of quartz, obsidian, jasper, chalcedony, silicious slate, etc. (Fig. 1—6).

The hind end of these stone arrow-heads is either slender and thus fitted to be inserted into the fore end of the shaft (Figs. 2, 3, 6), or they are devoid of such tang or shank (Figs. 1, 4, 5) and are connected with the shaft by means of a horny arrow-nock (Fig. 14) or by other means. The horny arrow-nock is coated with pitch at both extremities. Some arrow-heads wanting the tang are barbed (Fig. 5).

Many arrow-heads are made of bones or antlers. They are found from shell-mounds of various localities. There are many kinds as to shape—flattened and lanceolate (Fig. 9), long and conical (Figs. 10—13), unilaterally barbed (Figs. 16, 18, 22, 27), or bilaterally barbed (Figs. 19, 23, 25, 26, 30, 36), and they are tapering or narrow at the hind part with a coating of pitch to serve as the tang. The number of barbs varies much. In Fig. 23 four barbs on one side and three on the other side, and in Fig. 27 one incomplete and four complete barbs are found on one side only.

These barbed heads are abundantly found in many shell-mounds. They are about 40—60 mm. in length, of which about one half serves as the tang and is coated with pitch. They are thick and ellipsoidal or ovoid in cross-section. Almost all the barbed heads are made of stag-horn, I found among the rich collection of Mr. TAKASHIMA only one specimen made of bone (Fig. 36). This rare form was discovered in the shell-mound of Osozawa, Iwata-ken.

Some large specimens among these objects may have served as dart-heads or spear-heads, but as there is a fine graduation in size as well as in form, we cannot tell the difference from these heads only.

Fig. 15 represents a barbed, not-tanged arrow-head of stag-horn having a small perforation near the hind margin. One side of it, corresponding to the external side of the antler, is more or less convex, while the other side, corresponding to the interior of the horn, is more or less straight. This was found in the shell-mound of Togu near Sendai. Fig. 20 rep-

resents another form of arrow-head of similar type. It is thin, being made of the enamel of a boar's tusk, and has very sharp edges. It is about 1 mm. thick and has a small hole, about 1 mm. in diameter, near the hind margin, where a slight notch is found at the median line. This fine and rare specimen was obtained from the shell-mound of Shiizuka, Ibaraki-ken. These two specimens are in the possession of Mr. TAKASHIMA. Arrow-heads wanting the tang and made of other materials than stone are rather rare.

Many curious pointed rods, made of antlers, were discovered by Mr. TAKASHIMA in the shell-mound of Miyatojima, the largest of the famous pine-clad islets of Matsushima. Two of these rods are represented in Figs. 17 and 24. These rods vary from 60 to 90 mm. in length and they are curved a little and pointed at both extremities, one of which is sharper than the other. The convex side of these objects is cut or ground straight just below the comparatively blunt extremity. Thus this straight side makes an obtuse angle with the sharper extremity. The middle portion of these rods is coated with pitch on the straight side as well as on the concave side where we find traces of transverse lashing. In some rods the concave side is more or less hollowed, most probably to receive the lashing (Fig. 24). I am inclined to believe that these pointed objects were used as the apical portion of missile implements, such as arrows or darts. The straight side of these objects being apposed to a corresponding side of the shaft, they were lashed fast together, thus the comparatively blunt extremity serving as the point and the sharp extremity as the barb. Some of these objects were conjectured by MUNRO* to be fish-hooks.

Quite recently Mr. K. NAKAJIMA obtained two specimens of similar objects from the shell-mound of Kuwagasaki, Iwate-ken. They are about 88 mm. in length. One of them is thick and quadrangular in cross-section while the other is laterally compressed (Fig. A). The former (a) resembles more or less the original of Fig. 24 and has the anterior pointed part about 10 mm. in length, which is followed by a narrow stem of about 40 mm. The surface of the stem is more or less rough and is coated with

* Prehistoric Japan. 1908. p. 142.

pitch. There is no straight side at the stem. The barb is about 27 mm. long. The latter specimen (b) resembles the original of Fig. 17, but is longer, thinner and wants the straight side. The anterior pointed part is 20 mm. long, 9 mm. in breadth and 3 mm. in thickness. The stem is



Fig. A. Dart-heads of deer-horn,
From Kuwagasaki.

about 30 mm. long, 6 mm. in breadth and 4.5 mm. in thickness. On its external side many transverse scratches are found. The barb is nearly equal to the stem in length, breadth and thickness. On this specimen I find no obvious trace of any cementing substance.

There are double-pointed stone implements, which I think may have served as dart-heads, if not as arrow-heads. Fig. 7 represents one of these implements, made of a flaked stone. The implement measures 60 mm. in length, 10 mm. in breadth, and 4 g. in weight. It is more or less quadrangular in cross-section and tapers gradually towards both ends, being broadest in the middle. It was found by

Mr. SUZUKI near the source of the Shonai River, Yamagata-ken. Fig. 8 represents another implement of a ground stone, obtained by the same gentlemen near the mouth of the Yoneshiro River, Akita-ken. It measures 86 mm. in length, 10 mm. in breadth, and 10 g. in weight. Its cross-section is more or less round. From the shell-mound of Yoyama, a village near the mouth of the Tone River, Mr. TAKASHIMA obtained a similar implement, dart-head of a ground stone, measuring 103 mm. in length, 11 mm. in breath, 7 mm. in thickness, and 16 g. in weight. About the use of such double-pointed implements, previous writers have conjectured and discussed them as dart-heads, bait-holders or part of fish-hooks;

but as these implements are found in our country together with elaborate hooks, I believe it proper to classify them as dart-heads.

Fig. 28 represents a very interesting form of dart-head. It was found at Yoyama by Mr. TAKASHIMA. The implement is made from the tail-spine of a ray (*Trygon akajei*). The proximal portion of the spine is carved to a slender tang and coated with pitch. The spine is about 8 mm. broad, rather large, but as the distal portion is broken off its entire length is unknown.

A long, nonbarbed dart-head, made of a bone (Fig. 20), is very common and widely distributed. It is sharply pointed at one end, blunt at the other and often has a narrow neck near the blunt end. It measures about 100—120 mm. in length and 7 mm. in breadth. Its cross-section is mostly circular. The use of this implement for fishing was proved by the discovery of a skull of tai (*Pagrus major*) with the end of such implement thrust into its substance* (Fig. 126). The free portion of the dart-head outside the skull has been broken off. The dart was delivered by the right hand, from the rear, and it hit just the central part of the coalesced frontals. This remarkable skull was discovered from the shell-mound of Shiizuka, a village near the southern shore of Kasumigaura, a lagoon in Ibaraki-ken.

At present arrows or darts are no longer used for fishing in our country.

SPEAR-HEADS OR FIXED HARPOON-HEADS.

There are spear-heads of stone, antlers, or bone, and they are either barbed or nonbarbed. They vary in total length, from about 100 to 200 mm.

Spear-heads of stone are as far as I know nonbarbed. They are rather rare. One specimen in my collection from Niigata-ken is triangular in cross-section.

* KISHINOUE—Japanese Species of the Genus *Pagrus*. Journ. Fish. Bureau, Tokyo. Vol. X, 1901.

Though MUNRO states that he has not seen any spear-heads of bone, a report on the North Kurile Islands published by the Hokkaido Local Government in 1909 contains illustrations of such implements, both barbed and nonbarbed. A barbed specimen measures 123 mm. in length and 20 mm. in breadth and has a pair of barbs near the hind end and a small perforation in the flattened tang. A nonbarbed head measures about 180 mm. in length and about 30 mm. in breadth. From the porous appearance in the figure this object seems to have been made of the bone of a whale. It is probable that the implements described as above were used for the capture of marine mammals such as whales, porpoises or seals.

Besides the spear-heads of stone and bones, there are a great many spear-heads or fixed harpoon-heads made of antlers. Spear-heads of antlers are generally furnished with many barbs on both sides, and these barbs are mostly bilaterally opposite. As these implements have been made of the external hard portion of the stag-horn, one side of them is often convex while the other is flat or concave, the coarse and porous internal portion having been removed as much as possible. As antlers are rarely straight, large spear-heads made of them are naturally deflected more or less from the straight line (Figs. 29, 31, 34, 35).

An enormously large spear-head (Fig. 32), about 30 mm. broad and 16 mm. thick at the base or tang, was collected by Mr. TAKASHIMA from the shell-mound of Yoyama. This implement was carved out of the proximal portion of an antler and its root, the part inside the skin was utilized as the tang of the instrument. Though the anterior portion was broken away, there remain two pairs of barbs.

Some spear-heads have only one barb. Fig. 29 represents an example of such implements. The barb is found about midway between both ends. The implement is more or less rounded in cross-section and measures 140 mm. in length.

A spear-head found at Kamegaoka, Aomori-ken, on the coast of the Japan Sea, is remarkable in having two barbs, which are on two different planes, perpendicular to each other (Fig. 33). The proximal end is

blunt and there is a wide but slight constriction a little above the end. The implement measures 147 mm. in length and is in the possession of Mr. S. YEMI.

Generally speaking the spear-head made of antlers has a short tang, its length being $\frac{1}{4}$ — $\frac{1}{2}$ of the total length and in an extreme case even $\frac{1}{8}$, and the part is often found coated with pitch.

Holes, grooves, and protuberances in the spear- or harpoon-heads are believed by many archaeologists to be devices to retain the detachable head to the shaft, but these devices are sometimes used to make the lashing of the head to the shaft fast. Indeed some spear-heads with a coating of pitch on the base have exactly the same form as the harpoon-heads which from their structure are supposed to be detachable. The spear-head which has a perforation in the median line, anterior to the last pair of barbs (Fig. 37) cannot be thought to be detachable. The perforation in this case would probably be used to pass the lashing connecting the head to the shaft. This implement and other similar objects have been found in the north-eastern district of Hondo. Fig. 35 represents a curious spear-head from Osozawa in the collection of Prince NIJO. There is a pair of somewhat triangular lateral ridges just above the tang and these ridges are separated from the other parts by paired notches. The tang is coated with pitch, hence the ridges and notches would have served to fasten the lashing connecting the head to the shaft.

Barbs are sometimes long and sharp, sometimes short and blunt. In the bilaterally barbed specimen the number of barbs on both sides often differ very greatly. In one instance we count five barbs on one side and only two indistinct ones on the other (Fig. 31). In the case of many barbed specimens the most proximal barb or pair of barbs is found just above the base or tang. So far as I know the maximum number of barbs found on one side of the harpoon-head is nine (Fig. 34).

At present spear-heads are rarely used in fishing in our country and those now used have only one barb or one pair of barbs. Implements of this kind are employed for catching the fan-mussel (*Pinna*), devil-fish (*Octopus*), etc.

DETACHABLE HARPOON-HEADS.

Detachable harpoon-heads are either barbed, spurred, or barbed and spurred; and we find a great many varieties in form and structure. Moreover there are several devices in connecting the detachable heads to the shaft. The detachable form of harpoon-heads is used for the capture of big game which are not easily caught but struggle violently to escape. Generally these heads enter completely into the body of the victim and are connected with a line to the shaft which then serves as a float. Thus these heads are necessarily barbed, but mostly short. The connecting line is generally fastened at the middle, and when it is perpendicular to the plane of barbs the implements would be more effective than when it is in the same plane with barbs. All the detachable harpoon-heads are made of antlers.

HARPOON-HEADS WITH CONSTRICTIONS, PROTUBERANCES
OR THE LIKE.

These instruments are made of antlers and are bilaterally barbed and these barbs mostly symmetrical. The base or tang is rather short, $\frac{1}{3}$ — $\frac{1}{5}$ the total length and its surface is quite smooth. Often the base is broad at the anterior portion and, tapering gradually towards the free end, assumes the shape of a triangle or wedge (Fig. 38), or a cone (Fig. 46). In the harpoon-head represented in Fig. 38, the base is considerably broader than the stem, thus at the anterior end of the base, a pair of protuberances is formed. In a specimen illustrated in Fig. 46 a circular constriction between the stem and the base is very well marked. This specimen was collected at Miyatojima. Fig. 40 represents a harpoon-head from Osozawa. Its base is remarkably short and is hardly broader than the stem, from which it is separated by a narrow constriction. The original of Fig. 42 is a very large, bilaterally barbed harpoon-head, found at Yoyama by Mr. I. Ono. On one side barbs are more acute and more numerous than on the other, and on the side with more barbs we find a protuberance at the base,

Besides the types hitherto described there are some forms which have neither distinct constrictions nor protuberances, but whether they belong to the Stone Age or not is doubtful. This special type (Fig. B) comes from Hokkaido (Kunashiri, Poromushiri, and Shumshu). Harpoon-heads belonging to this type are very finely made, gradually flattened and more or less trenchant at the margin of the anterior part, which is armed with one or two pairs of barbs. The tang is long, and more or less spindle-shaped, the bulbous portion serving to retain the connecting line. The length of these heads varies from 100 to 170 mm.

Mr. TAKASHIMA has two remarkable forms of harpoon-heads, in which the posterior part or the base is bent to one of the barbed sides. There is no marked boundary between the shank and the base as in the last mentioned type. One of these harpoon-heads (Fig. 41) was discovered in the shell-mound of Mitsusawa and is of fine workmanship. This specimen is long and flat, measuring 130 mm. in total length, 140 mm. in breadth, and 3 mm. in thickness. It is provided with two pairs of strong barbs, which are not exactly symmetrical on both sides. On the concave side of the base we find a slight protuberance or a shoulder-like prominence which served probably to retain the line, connecting the head to the shaft, when the former is detached.

Another form (Fig. 47) was found at Miyatojima. It is 100 mm. long, 11 mm. in breadth at the base which is more or less rounded in cross-section. The implement is armed with two pairs of barbs, the anterior pair of which is pointed while the posterior pair ends in a line. This is a very peculiar and seemingly absurd structure, but when thrust into the body of an animal such a barb would rarely come loose. In this implement there is neither groove nor knob at the base, but as it is swollen at the middle, a line tied anterior to it would not fail to connect the detachable head to the shaft. The portion behind the



Fig. B.
Harpoon-head
of deer-horn.
From
Kunashiri.

second pair of barbs is more than twice longer than the portion before the pair. The base or tang is bent to one of the barbed sides as in the last case. From the smoothness and acuteness of the base which is swollen at the middle I am inclined to believe that this form was also detachable.

The two last mentioned objects have a resemblance more or less to the curious pointed objects represented in Figs. 17, 24 and described as arrow-heads or dart-heads, especially to a similar object, without coat of pitch, found at Kuwagasaki.

PERFORATED HARPOON-HEADS WANTING SPURS.

They were made of antlers and have a strong barb or barbs. The base of these implements is rather short, slender, and more or less rounded in cross-section. The perforation is made either perpendicular or parallel to the plane of the barb or barbs, and is found near the middle where the material is generally most thick and broad. Notwithstanding this precaution these implements are often damaged from the perforation (Figs. 43, 44). They are rather rare but manifold in structure, so that I found no two alike. So far as I know they are known from shell-mounds of the north-eastern district of Hondo only.

The most simple form I examined is illustrated in Fig. 43. It has only one large barb and an aperture in the median line. The proximal portion next to the hole is broken. This form was found at Osozawa, Iwate-ken.

Another form with only one barb (Fig. 39) was found in the same locality, but its aperture is in a special knob which is situated on the side opposite to the barbed side. The knob is broad, sharply edged at the abaxial side and is pointed at the abaxial, proximal corner, so that it would have served as a barb too. This specimen has a squarish cross-section at the stem. Harpoon-heads with a perforation in a knob were found in Europe and America, but they differ from the present form by having the barb and knob on the same side.

Fig. 45 represents a harpoon-head from the shell-mound of Miyajima. It is about 90 mm. long, 10 mm. in breadth, and 6 mm. in thickness. It is armed with two barbs on one side and only one at the other. The perforation is found at the median line of the stem and near the origin of the base, which is $\frac{2}{9}$ of the total length and is narrower than the main stem.

Fig. 44 shows a broken harpoon-head, damaged at both ends. It was obtained by the author from a shell-mound at Numazu, near Ishinomaki, Miyagi-ken. The anterior end and the portion posterior to the perforation are broken off. The actual length of this broken specimen is 60 mm. It is armed with a long triangular blade at the anterior end and a pair of narrow barbs. It is nearly flat on one side and keeled at the median line of the other side. The flat side coincides with the inner or axial side of the antler, while the keeled side is the cortex. The stem between the triangular blade and the paired barbs is irregularly pentagonal in cross-section, while the stem posterior to the barbs is rounded. The paired barbs are quadrangularly pyramidal. The perforation of about 4 mm. in diameter is found in the plane of barbs. Along the posterior margin of the triangular blade and on the lateral sides of the stem just before the paired barbs, we find two linear grooves. I cannot believe that these grooves either served to receive poison or to facilitate the flow of the wounded animal's blood. They seem to be simply ornamental, as the uncivilized people often decorate their fishing apparatus with unnecessary ornaments.

In an iron harpoon-head, obtained at Karafuto (Saghalien) we find a somewhat similar construction as in the last mentioned instrument. They are equal in having one side flat, the other side keeled and the stem angular in cross-section.

HARPOON-HEADS WITH ONE OR MORE SPURS.

They were made either of staghorn only or of stone and staghorn. They were generally made of the terminal part of a horn or its tynes and their axes correspond with the axis of the horn or tynes. These imple-

ments are chiefly and not seldom found in the north-eastern district of Hondo; Kuwagasaki, Osozawa, Nakagawa, Numazu (near Ishinomaki) and Miyatojima. Only one simple form of this kind (Fig. 57) was found at Yoyama. Thus so far as I know the last named locality is the southern limit of distribution of the spurred harpoon-heads.

Generally the spurred harpoon-heads have two holes, transverse and longitudinal. The former is the line-hole, made through the body and is for a string connecting the head with the shaft. The latter is wide, short, terminates bluntly and serves as a socket for the anterior end of the shaft. The transverse hole is finished by boring from both sides, so that the hole is very narrow in the middle. The longitudinal hole is nearly conical and found at the posterior end. The hole or socket is to fit to the anterior end of the shaft or the intermediate piece or fore-shaft if such one was used and of course does not reach the level of the transverse hole. The socket is about 10 mm. in diameter and 5—13 mm. in depth. I have not yet found a line groove in these harpoon-heads.

The simplest form of the implement found at Yoyama is 57 mm. long, nearly circular in cross-section, with a spur of about 20 mm., bifid at the posterior extremity. Similar specimens (Figs. 50, 54) were found at Miyatojima and Numazu near Ishinomaki. From the latter locality I have another specimen of similar form with a trefid spur. Recently a specimen of this type was obtained by Mr. NAKAJIMA from the shell-mound of Ozuke near Kuwagasaki. The spur is bifid, divergers towards the distal end, and is rounded in cross-section.

A large number of mostly larger and more developed forms are found at Osozawa and adjacent places. Among them the evolution in the shape of the apical part and the spur is instructively seen (Figs. 53, 58—60).

The former becomes provided with a barb or barbs on the side opposite the spurred side. Then the barb is formed on the spurred side also and thus the apical portion becomes laterally flat, specializing the blade portion from the other.

In most specialized forms a stone blade is found in a deep slit or kerf at the apical part (Fig. 29). The stone blade is cemented to the slit with pitch. In these forms the blade-slit or rather the stone blade itself is protected by a pair of high walls or long processes, and each of these processes is bordered at the proximal part with a groove. As the outer surface of these processes is at the distal part rough and coated with pitch we are inclined to believe that the stone blade has been fixed in its position by some lashing too. The spur becomes broader and more deeply divided into two, three or even four spurs. The barbs may be unilateral or bilateral and their number and form are subject to variation. They appear first on the side opposite the side of the spur. In a complicate form we find two or three pairs of barbs besides the apical portion.

It is very interesting to note that the blade, barb, spur, and transverse hole are all in the same or nearly the same plane. Such implements used in other countries as well as in our country at present have the transverse hole and the spur in different planes, perpendicular to each other, while the blade and barb are in the plane of the transverse hole.

An implement of this kind requires much time and labor to make, but it is so easily broken that the prehistoric people lashed the broken pieces together with the addition of pitch, and often they made a cut or notches at the broken part to secure the lashing (Fig. 54).

Two very singular forms of the spurred harpoon-heads (Fig. C) were recently discovered by Mr. KICHIBEI NAKAJIMA in the shell-mound of Kuwagasaki and were kindly sent to me by the finder. They are very simple and differ from those already found in wanting the transverse hole or line hole but having a transverse groove round the implement at the level just a little above the distal end of the longitudinal hole or the socket for the shaft. At the transverse groove we find a coating of pitch, 6—10 mm. broad. Thus we see that the string connecting the head and the shaft was tied round the groove and was cemented with pitch. In both specimens the transverse groove is found at a distance of about 10 mm. from the proximal end.

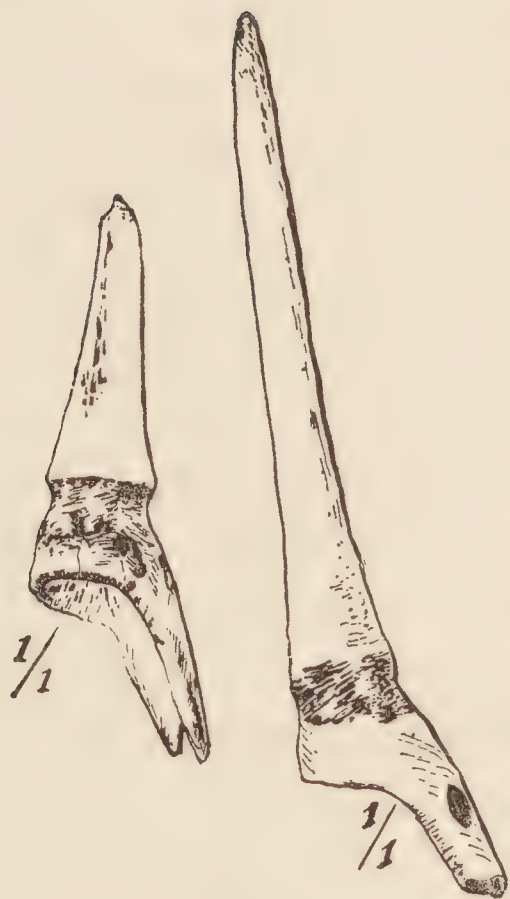


Fig. C.
Harpoon-heads of antler.
From Kuwagasaki.

One specimen is 35 mm. long with a bifid spur of about 15 mm. and its diameter at the proximal end is 12 mm. The other is 68 mm. long with a perforated spur of 12 mm. and its diameter at the proximal end 11 mm. The perforation in the spur is remarkable. I cannot understand for what purpose it has been made. It seems to have been bored from the axial side, as its diameter is larger at the side. In both specimens the anterior part is conical.

In a manuscript written some thirty years ago on the fishing industries of Miye-ken at that time, illustrations of harpoon-heads are given. Each of these implements has a small perforation near

the distal end of the spur and the perforation is stated to serve sometimes as a line-hole.

A detachable form of harpoon-heads commonly used at present in our country has the longitudinal hole only. The head is gradually swollen towards the proximal end and the distal end is sagittate, furnished with a pair of long, pointed barbs. The string connecting the head to the shaft is tied just anterior to the distal end of the longitudinal hole, as in the case of harpoon-heads from Kuwagasaki, but there is no groove for the string. The spur is simple and is a little reflexed abaxially.

These spurred harpoon-heads seem to have been used for the capture of large fish such as tai, tunny, sword-fish, etc., and also of marine mammals, such as seals, porpoises and whales.

PRONGS OF FISH-SPEARS.

Instruments represented in Figs. 48, 49, 51, 52, 55, 56 represent prongs or tines of fish-spears. They are made of staghorn and are pointed or barbed at one end and are furnished at the other end with notches or ridges on one side and an obliquely straight surface at the opposite side. I think that the side with notches or ridges is the abaxial side, while the side with straight surface is axial. Generally these prongs are more or less curved towards the axial side. They are 70 to 160 mm. in length, 5 to 20 mm. in breadth and are generally rounded in cross-section, but the prongs represented in Figs. 49, 55, 56 are more or less flattened, especially near the proximal end.

The barb is found either on the axial, abxial or both sides and in one case (Fig. 55) the barb seems to have been made from a material, different from that of the prong. Most probably the point and the barb were made from a piece of stone. The prong has a longitudinal groove, 12 mm. long, 3 mm. broad, on the axial side of the proximal end to receive a pointed piece of another hard material and is coated with pitch in the groove as well as on the outer part.

The proximal end of these prongs is also coated with pitch (Figs. 48, 51, 55), nearly to the level of the distal end of the straight surface. The number of notches or ridges at this part is generally one or two, but in one specimen from Osozawa we find many (Fig. 49).

Probably two, three or more of these prongs were lashed to one end of the shaft, and were used for the capture of some shore fish such as tai, perch, grey-mullet, etc. The geographical distribution of these instruments is much restricted. So far as I know a dozen of them were found in Miyatojima, one at Osozawa and another piece at Hosoura.

Mr. MUNRO takes some of these prongs for hooks, but from the long and nearly straight shape and mostly thick and large size I cannot agree with him. In the regions where these objects were found, very elaborate hooks of varying sizes were also found at the same time. Thus we

cannot believe that the same people would have used such implements as hooks.

In the fish-spear now used the barb is mostly found on the axial side of each prong. Prongs of a spear for shell-fish, sea-urchins or some other fish, however, have no barb at all. Prongs used at present are about 200 mm. in length, but some small examples measure about 50 mm. only. Larger and heavier prongs are used in deeper waters.

Hooks.

There are various kinds of hooks found in different districts. All of them are formed of staghorn and I have not yet found any hook made of other material, such as stone, shell, wood, etc., nor did I find any hook made from two different materials. Many hooks are very elaborate and may be used effectively even at present and nearly all prehistoric hooks hitherto found in our country seem to have been used with some bait. Up to the present time prehistoric hooks were found chiefly in the north-eastern district of Hondo. It is rather curious and noteworthy, that hooks from the northern district near Sendai are more advanced than those from the district near Tokyo. This fact may be explained by the longer and later occupation of the prehistoric tribe in the former region than in the latter. The same fact is noticed in potteries and other instruments made of bones and staghorn.

For fastening a line to the hook, the upper end of the latter is provided with notches, grooves, ridges, perforations, etc.

Very recently Mr. KICHIBEI NAKAJIMA sent me a broken piece of an implement of stag-horn, which seems to be the upper extremity of the stem of a fish-hook. However, it is quite different from the similar part of other hooks already discovered, the extremity of the stem being bent a little and the bent part grooved on both sides. In the groove I cannot find any trace of a pitch-like substance, but near the lower end of the groove and at the junction of the upper extremity and the stem there is a very faint streak of black color. The peculiar structure

of the implement reminds me of some fish-hooks from California* and a fish-hook of reindeer-horn from the Norwegian part of Lapland.† The length of the broken piece is about 30 mm., diameter 4 mm., and the length of the bent part 10 mm. It was found at Kuwagasaki.

BARBED HOOKS.

Prehistoric hooks hitherto discovered in our country have mostly a barb or barbs on the outer side of their stem. Some hooks are barbed both on the inner as well as the outer side. On the inner side we find only one barb, never more, while on the outer side we find often three barbs. From the shell-mound of Kuwagasaki Mr. NAKAJIMA obtained some hooks which have no outer barb but one inner barb only.

The outer barb is mostly found near the boundary between the apical part and the base or curved portion (Figs. 61, 64, 70, 71). This is the case when there is one barb only, but when there are two or three outer barbs, they are found between the point or apex and the curved portion (Figs. 63, 65, 66, 69, 75).

The axis or stem is generally straight and the curved portion or base is considerably thicker than the other parts, especially in the vertical direction (Figs. 63, 65—67, 69—71, 75). Notwithstanding this precaution, hooks are broken generally from the curved portion when they are subjected to a great strain (Figs. 62, 72).

Fig. 61 represents a very primitive form of our prehistoric fishing-hooks. It was obtained from Kubiri, a village near Uruga, Kanagawa-ken. It measures 25 mm. between the pointed end and the outer barb, while the stem is about 45 mm. long. At the upper end of the stem there is a notch on the external side for making the whipping to a line fast. The cross-section of the stem and the curved portion is rounded, while that of the apical portion is more or less crescent-shaped, convex towards the outer side. There is only one external barb, nearly on the

* Rau-Prehistoric Fishing, pp. 129-133.

† Ibid. p. 72. Fig. 93.

curved portion. This peculiar position of the barb reminds us of the hooks of Eskimos and those from Greenland, described and illustrated in RAU's paper on prehistoric fishing in Europe and America. Thus the hook from Kubiri has a rather clumsy appearance as a whole. This specimen is kept in the Anthropological Institute of the Imperial University of Tokyo.

Fig. 64 represents another primitive hook, found at Kashiwai, Okashiwa village, Chiba-ken and now in the possession of Mr. S. YEMI. It is quite perfect and comparatively well-made. It measures about 60 mm. in the stem and about 30 mm. in the base. The stem is nearly straight, curved a little towards the inner side and has an angle of about 100° with the apical portion. The material is very thick at the angles. The upper extremity of the stem is smooth and rounded and a little below the end there is a groove round the stem. The apical portion is conical and ends in a sharp point. An external barb is found at the angle of the base with the apical portion and the barb is separated from the base by a groove from one side only as is shown in the figure. A similar groove is found between the apical portion and an internal barb in some metallic hooks now used in some out-of-the-way places.

Mr. YEMI has another hook of somewhat similar form, collected from the sea-shore of Katsuura, a small town on the east coast of Chiba-ken. The hook is much corroded, especially at both extremities. It is nearly equal in size with the last mentioned specimen. The stem and the base are rounded in cross-section and at one end of the base we find a vestige of an external barb.

A large number of hooks with an external barb have been found in the shell-mound of Yoyama and are in possession of Messrs. I. ONO and T. TAKASHIMA. These hooks are characterized by their large size, long and straight stem and well-developed upper extremity. Their make is rather elaborate. The majority of these hooks has the stem from 90 to 120 mm. in length and the smallest hook I observed measured 60 mm. in the length of the stem. The form is nearly of one pattern. The stem, base and apical portion are nearly straight and the stem and the apical

portion are almost parallel to each other, while the base makes nearly right angles with them. Of these three parts the base is shortest and thickest.

Fig. 70 shows a fine and complete specimen from the collection of Mr. ONO. The length of the stem measures a little more than 90 mm., its diameter about 8 mm.; the base is more than 30 mm. in length and 15 mm. in diameter; and the apical portion about 40 mm. in length and nearly parallel to the stem. The barb is found on the external side and near the boundary between the apical portion and the base. Near the upper end of the stem a ridge is found round the stem.

In another specimen from Yoyama, measuring about 100 mm. in the stem and about 20 mm. in the base, the upper end of the stem is nearly flat at the internal side, while there are two transverse ridges on the external side to ensure the lashing of a line. A remarkably well-developed example of this kind of a hook is represented in Fig. 74. In this specimen the upper end of the stem is considerably wide, as it is about twice as broad as the diameter of the remaining portion of the stem. On the internal side of this broad part we find a shallow but distinct longitudinal groove, while on the external side there are two well-developed transverse ridges. The upper ridge is narrow and steep, while the lower is broad and rises gradually from the former. Between these two ridges we find naturally a transverse groove. From these longitudinal and transverse grooves we may conjecture more or less the thickness of lines then used. The breadth of these grooves varies from 1.5 mm. to 3 mm. Thus we see that lines then used were comparatively very thick. Moreover from these structures of hooks we understand that a line was apposed to the internal side of the stem of a hook and then it was whipped with another line at the transverse groove.

In a few forms of hooks from Yoyama we find the barb very well-developed and turned out externally, nearly making right angles with the tangent at the boundary between the apical portion and the base.

Many half-made hooks were also dug out from the same locality. These together with similar objects from Miyatojima (MUNRO—Pre-

historic Japan Fig. 50, p. 142) and other places explain more or less by themselves the process of hook-making of the time. We consider that the process was as follows:—

At first an antler was split into two longitudinal halves. Then a rough form of a hook was carved out from these pieces of horn. Hooks were cut away from the mother piece of horn at this stage or when nearly finished. Thinning of the material and chiseling a barb or barbs near the apical portion and carving ridges or notches at the upper end of the stem were next performed. Scars left by chiseling are well observable, generally in very fine and mostly paralld zigzag lines. Lastly a pretty careful polish was given and thus the process of manufacturing hooks was at an end. Of course the long axis of the hook corresponds with that of the material, and as the hard and compact part of the material is curved the base of a large hook is generally curved to either side:

Mr. TAKASHIMA obtained two complete hooks from the shell-mound of Shiizuka. One of them is represented in Fig. 71. Having a long stem, these hooks are nearly equal in form to those found at Yoyama, a place about 30 km. distant. Besides the form, the size is almost alike. In the specimen represented in the figure we find two slight ridges at the upper end of the stem, while no special structure is found in the other specimen.

In 1909 I found a fine hook (Fig. 65) in the shell-mound of Numazu near Ishinomaki. It differs greatly from the hooks already described. The stem is nearly straight at the upper half but it is gradually bent with a fine curve at the lower half and passes to the base. Two transverse ridges are distinct on the external side of the upper end of the stem. The lower ridge is more prominent. The stem is angular in cross-section. The base is short, curved and has no distinct boundaries. Its cross-section is elliptical, as it is compressed vertically. The apical portion is considerably long and its point nearly touches the level of a ridge at the upper extremity of the stem. Three barbs, almost equal in size and equidistant from each other are found on the external side of the apical portion.

A large number of hooks were found in Iwate-ken—Osozawa, Nakazawa, Hosoura and Kuwagasaki, and they are in the possession of my laboratory, the Anthropological Institute of our University, Prince NIJO, Mr. TAKASHIMA, etc. Generally the hooks are small, 20—40 mm. in length of stem, and they are armed with an internal barb besides an external barb or barbs (Fig. 63, 66, 67, 69). A few forms are without the internal barb (Fig. 75.) and some others want the external barb.

Fig. 66 represents a large hook, nearly complete and armed with four barbs. It was found at Osozawa. The actual length of the stem which seems to have lost the uppermost extremity is about 50 mm. and it is about 6 mm. in diameter. On the internal side of the apical portion, there is a very broad barb which originates near the base. On the external side there are three barbs, the lowest one of which is found just near the junction of the apical portion with the base.

A similar but smaller hook found in the same locality is represented in Fig. 69. In this specimen we find also one internal and three external barbs. The apical portion is very long, reaching to the lower boundary of the upper extremity of the stem. The latter part is considerably long and tapers gradually to a point at the end, while the lower boundary is indicated by an external protuberance.

Figs. 63 and 75 also represent hooks from Osozawa. The former is armed with one internal and two external barbs. Its stem is more or less curved internally and the upper extremity of the stem is provided with an external knob and moreover it is coated with pitch. The coating of pitch at the upper extremity of the stem is rarely found and I have never seen it in hooks from the Kwanto-district, *i.e.* near Tokyo. The original of Fig. 75 is the smallest barbed hook ever found in shell-mounds in Japan. It is about 40 mm. in total length, measured along the curvature.

On a broken stem of a hook from Osozawa and another similar stem from Nakazawa, we count three external knobs on the upper extremity. Of these knobs the lowest one is most prominent and the uppermost one is most depressed. We see that the form and structure of the upper

extremity of the stem is greatly variable and often they are not constant even among the hooks from the same locality.

Fig. 67 shows a fine hook discovered by the author on the shell-mound of Hosoura. Its apical portion is very long, slender and curved internally. It is armed near the lower boundary with one internal and one external barb. They are situated nearly opposite each other. The base is broad and laterally compressed. The stem is nearly straight and has two small external knobs at the upper extremity.

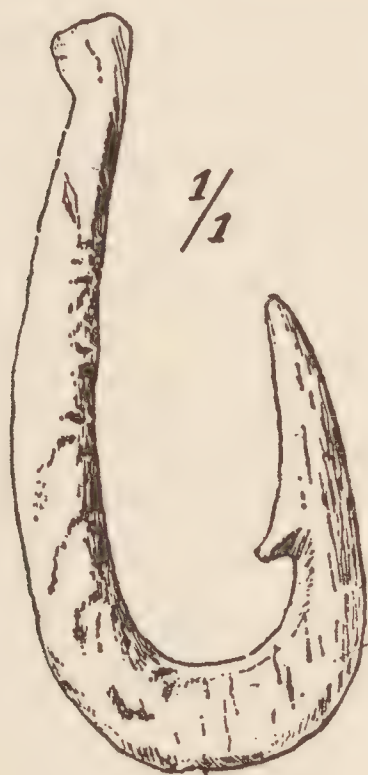


Fig. D.
Fish-hook of antler.
From Kuwagasaki.

A fine and perfect hook of much advanced form was of late discovered by Mr. K. NAKAJIMA in the shell-mound of Kuwagasaki (Fig. D). Its total length is about 110 mm. This hook is remarkable in having an internal barb only which is found rather nearer the base than the pointed end of the apical portion. The stem is long, curved a little internally and becomes gradually broad from the upper extremity towards the base, though the thickness is nearly uniform. The upper extremity of the stem is armed with an external knob which is more or less triangular in outline when seen from the side.

Two other specimens of hooks with an internal barb only were obtained by the same gentleman from the same locality. Unfortunately they are not complete, but their general form may clearly be understood, and both of them differ from the preceeding hook and from each other also. One of them (Fig. E) wants the upper half of the stem and the pointed end of the apical portion. The remarkable points in the structure are the long and sharp barb and the very broad but short base, pointed at the lower margin. This peculiar structure of the base is also found in prehistoric hooks of Europe (KRAUSE—Vorgeschichtliche Fischereigeräte und neue



Fig. E.
Fish-hook of
antler.
From
Kuwagasaki.

Vergleichsstücke, Fig. 330) and America (Rau—Prehistoric Fishing, Fig. 189, p. 127). The apex of the barb passes beyond the midway between the stem and the apical portion. In this hook it is also remarkable that both lateral sides are quite flat, parallel to one another and the comparatively narrow borders between them are rounded. The other specimen (Fig. F) is very remarkable in form and structure. The most striking point is found in the structure of the upper extremity of the stem. That part is laterally compressed, broader than the rest of the stem, and has a round hole and a notch. The diameter of the hole is 3 mm. The stem is long and curved internally. The base is thick, rounded in cross-section, and is pointed at the lower margin near the junction with the stem. The end of the apical portion is broken and an internal barb is found at some distance from the base.

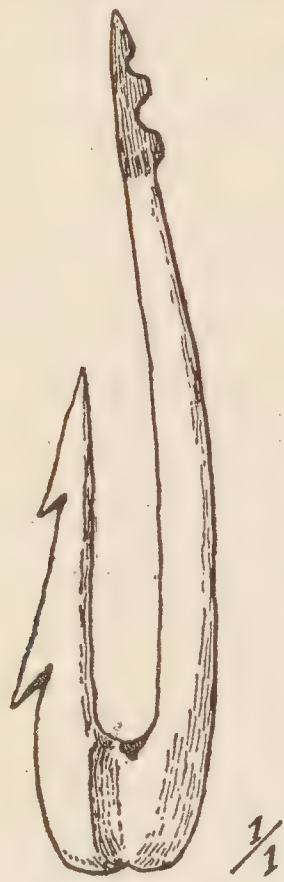


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Fig. F.

Fish-hook of antler.
From Kuwagasaki.

It is noteworthy that the internal barb found in hooks from Kuwagasaki is rather widely separated from the base. In this respect they are quite alike the hooks now in vogue.



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Fig. G.

Fish-hook of antler.
From Kamegaoka.

Recently a very fine hook from Kamegaoka, a village on the coast of the Japan Sea, Aomori-ken, was added to the collection of Prince NIJO (Fig. G). This is the only primitive hook from the coast of the Japan Sea and differs from other hooks in many points. The stem is very long, a little curved internally and is rounded in cross-section. The apical portion is also very long, nearly straight, parallel to the stem and is provided with two external barbs. The base is very broad vertically, but exceedingly short, so that the distance of the stem and the apical portion is about 1/11 the length of the former and about 1/6 that of the

latter. On one side of the base we find a broad vertical groove which was made probably by scraping off the porous portion of the material, the antler. Besides this groove there are two notches at the lateral edges of the upper surface of the base and a transverse groove at its lower surface. These notches and the groove seem to have served for lashing something, probably some bright colored material for alluring fish. The upper extremity of the stem is armed with three external knobs and is coated with pitch.

Comparing numerous hooks hitherto found in different parts of Japan, we observe the gradual changes which took place in the form of hooks. The most primitive form is curved somewhat angularly and has an external barb near the junction between the base and the apical portion. Then the number of the external barb increases to two, then three and at this time an internal barb makes its appearance. At first the position of the barb, whether it is internal or external, is as low as possible, and then it is found to shift gradually towards the pointed end. The number of the external barbs afterwards diminishes again and at last they disappear entirely, leaving the internal barb only. The stem and the apical portion are at first straight, but they become afterwards curved internally.

The form of the barbed hooks now used in our country is manifold and cannot be discussed in a few lines, but all of them have only one barb on the internal side and rather near the apex. Their size measured from the apex to the upper extremity of the stem along the curve varies from 9 to 300 mm. In the hooks made in some districts on the coast of the Japan Sea and in Formosa the barb is formed by filing an oblique notch from a lateral side as we see in the hook represented in Fig. 64. The upper extremity of the stem of hooks is flattened in the plane of the hook or in a plane perpendicular to that of the hook and is provided with a pair of lateral protuberances or a groove or grooves on either one of the lateral sides, on both sides, or on the external side. This part of the hooks is rather more simple at present than in the Stone Age. Large hooks now used are often eyed, that is they are perforated at the upper extremity,

but we found only one such eyed specimen among our prehistoric hooks. The larger barbed hook from Kuwagasaki represented in Fig. D. is quite similar in size and form to a hook now used in a tunny long-line.

HOOKS FROM MIYATOJIMA.

Many large hooks were discovered in the shell-mound of Miyatojima by Mr. T. TAKASHIMA, but unfortunately they are all broken and want the apical portion (Figs. 62, 72, 73), so that we cannot judge whether they were barbed or not. From Fig. 73 we see that at least the original of this figure is not barbed near the lower boundary of the apical portion.

A fine curve, peculiarly shaped upper end of the stem, comparatively thick stem with a roundish cross-section, and generally stout structure characterize Miyatojima hooks. Moreover all of them have a coating of pitch at the upper extremity of the stem, which is triangular in a side view, and conically pointed at two points. Such peculiar structure of the upper end of the stem is found in hooks made of molluscan shells and discovered on the Santa Cruz Islands, California (RAU—Prehistoric Fishing, Figs. 207—209, P. 133). Generally the coarse porous portion of the antler is more or less found at the base, but in some hooks from Miyatojima the coarse portion is also found near the upper end of the stem, as they are large and much curved (Fig. 72). The base is also rounded in cross-section.

More or less similar hooks, also wanting the apical portion, were recently dug out by Mr. G. YENDO from the shell-mound of Numazu near Ishinomaki.

UNBARBED HOOKS.

Unbarbed hooks are rare and at present they are known from Iwate-ken only, Osozawa and Kuwagasaki. So far as I know three perfect specimens were obtained from the former locality by Mr. K. NONAKA, and three from the latter by Mr. K. NAKAJIMA. These unbarbed hooks

are generally small in size and very finely made, so that we are inclined to believe that they have been made in later periods than the barbed hooks.

Fig. 68 represents a hook from Osozawa and it measures about 36 mm. in length along the curvature and only 2 mm. in diameter. This apical portion is directed rather externally and is moderate in length, reaching about the middle of the stem. The distance of the pointed end of the apical portion from the stem is nearly equal to the length of the former. The stem is nearly straight and its upper extremity is provided with two external knobs. In the other two specimens the upper end of the stem is pointed and is provided with only one external knob.



Fig. H.
Fish-hook
of antler.
From
Kuwagasaki.

Fig. H. represents an unbarbed hook from Kuwagasaki. It is about 40 mm. in total length. The apical portion is very long, its length being contained a little less than one and half times in the stem. The distance of the pointed end of the apical portion from the stem is contained a little less than one and half times in the former. The stem is nearly straight, gradually increases its diameter towards the lower boundary and has an external transverse notch near the upper extremity. The base is very broad, thick and more or less pointed at the lower margin as in the case of Fig. E.



Fig. K.
Fish-hook of antler.
From Kuwagasaki.

Fig. K. illustrates another unbarbed hook from Kuwagasaki. It is flat on one side and convex on the other. The inner margin of the hook is straight and perpendicular to the plane of the hook. The total length is about 75 mm. The upper end of the stem and the pointed end of the apical portion are more or less curved internally, and at these parts the inner margin is slightly rounded. The base is broad or laterally compressed. The stem increases its diameter towards the lower end and is provided with an external knob. The make of this hook is rather rough.

A large and rather peculiar form of the unbarbed hook from Kuwagasaki is shown in Fig. L. The hook is 75 mm. in total

length. The stem is nearly straight, rounded in cross-section, and has an outward protuberance at the upper extremity. The base is thickened,



rounded and is slightly pointed at the middle of the lower margin. The apical portion is very short and is pointed abruptly from the base. Probably this hook was at first barbed, but being broken at the apical portion, the portion was renewed by carving the broken end sharp.

At present unbarbed hooks are generally used for hand lines or in angling with a rod, and similar hooks of the Stone Age would have been used in a similar manner, probably for the capture of some shore fish or pelagic fish, as bonito.

Fig. L.
Fish-hook of antler.
From Kuwagasaki.

THE LANDING HOOK OR GAFF.

Only one fine, large specimen is known. It was obtained from Kamegaoka and is now in the possession of Mr. Shitomi Sato. He has given me drawings of it, one of which I have reproduced in Fig. 76. The hook is made of horn, measures about 130 mm. from the tip to the other end and is bent in the middle at an angle of about 90°. The pointed end is armed with one internal and one external barb. The former is nearer the pointed tip than the latter. At the proximal end we find five transverse grooves on one side and a shallow longitudinal groove on the other side and this part is coated with pitch. The side with the transverse grooves corresponds to the cortical surface of the antler. From this we conjecture that a shaft was apposed on the side with the longitudinal groove and was firmly whipped with a string over and round the transverse grooves. At present gaffs have no barb generally.

Only those which are laid in the bed of a river awaiting the approach of a salmon are sometimes provided with an internal barb.

Sinkers.

We often find many objects which appear to have been used as weights or sinkers in fishing implements. It is, however, very difficult, almost impossible, to prove that these objects have really been used as sinkers or not. We simply take them as such, as they have a close resemblance to sinkers used at present and as many of them are found in shell-mounds. There is a great variety of these objects in size, shape, material, etc. They may roughly be classified into notched or grooved stone-sinkers, grooved clay-sinkers, perforated clay-sinkers and sinkers made of potsherds.

It is remarkable that many sinkers are found, as far as I know, in sites, different from those where hooks, fish-remains, etc., are abundantly found, and we understand that fishing villages were from remote times divided into those where line fishing predominated and those where net fishing prevailed. Sinkers are mostly found on shores of inland waters and inlets.

Notched stone-sinkers. Generally discoidal pebbles, elliptical in outline are selected as material. Notches are either flaked or ground and they are mostly found at the end of the longer diameter of sinkers (Figs. 89, 92, 94). Sometimes notches are also found at the ends of the shorter diameter (Fig. 93). When notches are ground they are rather narrow, ordinarily 1—3 mm. in breadth. These sinkers are about 20—90 mm. in breadth, 30—120 mm. in length, and 4—320 g. in weight.

Grooved stone-sinkers. Generally roundish or ellipsoidal pebbles are selected as material. These sinkers are mostly larger than notched sinkers and a groove is chipped round the sinker along the longer diameter (Figs. 90, 91, 95). Sometimes another groove is chipped crosswise to the first groove (Fig. 81). Fig. 88 shows a large specimen obtained from Tomariro, Karafuto. In this specimen the longitudinal groove does not

reach beyond the transverse groove. This sinker weighs about 1150 g. Fig. 95 represents a somewhat peach-shaped pebble with a chipped longitudinal groove obtained from the same locality. It weighs about 1250 g. The original of Fig. 90 seems to have been shaped quite artificially. It is more or less cylindrical and has a broad longitudinal groove on comparatively broader sides. It is damaged at one end and was found by Mr. S. SUZUKI at Onodai, Akita-ken. A discoidal form of grooved stone sinkers is shown in Fig. 91. It is small and is damaged at one end. The groove is very narrow and seems to have been made by grinding.

At present stone sinkers are still largely used—in gill-nets, dredges, lines, anchors, etc. For these purposes, fishermen collect pebbles or stones of proper shape and size or work them more or less to suit the purpose better. For nets and lines notched or grooved stone sinkers are rarely used. For nets natural pebbles are used as they are, or enveloped with straw and tied to the ground rope. For dredges generally artificially shaped stones, conical or pyramidal are used. Large stones weighing about 1.5—2.4 kg. are used for anchors or killicks. In such cases natural or artificially shaped stones of a convenient size are selected and often they are worked more or less to fit the wooden parts well. In Aomori-ken I saw a killick which was weighted with a round stone about 200 mm. diameter. The stone was perforated at the centre and the shank of the killick passed through the perforation.

Grooved clay sinkers. They are generally small in size, globular or ellipsoidal in form and blackish or brownish in color, as they are baked. I have not yet found any glazed sinker. So far as I know there is only one sinker which has only one groove (Fig. 78). The groove is at the narrower margin of the more or less flattened ellipsoidal body. This sinker was obtained from the shell-mound of Fukuda, Chiba-ken, by Mr. TAKASHIMA. In other specimens there are at least two grooves crossing each other at right angles (Figs. 77, 80). In a globular sinker I found three grooves crossing each other perpendicularly (Fig. 80). These sinkers weigh about 10—30 g. This kind of sinkers is not used in our country now. A small elliptical sinker was of late

collected by Mr. NAKAJIMA at Miyako. It has two grooves crossing each other at right angles. The groove running around the longer meridian is about 4 mm. deep and 3 mm. wide. The other groove is about 1.5 mm. deep and 2.5 mm. wide. The longer axis of the sinker is 27 mm.

Perforated clay sinkers. They are rather common and though many of them are clumsy, some are very finely made, scarcely differing from those now used. They are either spheroidal, cylindrical or spindle-shaped and generally brownish or reddish in color (Figs. 79, 82—84, 86, 87). Their size, weight and diameter of the perforation vary very much. They seem to have been bored through the median axis with a slender stick when they were still soft. Hence in some sinkers the border round the perforation remains more or less elevated (Fig. 86). In some other specimens borders round the perforation are made more or less flat and neat (Figs. 79, 82, 87). Spheroidal sinkers are very numerous and their diameter is mostly 20—30 mm. Cylindrical and spindle-shaped sinkers are rather few in number. The former shape is comparatively large and heavy while the latter is generally small, weighing two or three grammes only. The diameter of the perforation in clay sinkers varies from 1.5 to 15.0 mm.

At present cylindrical sinkers are used in drag-seines, trawls, etc., while small fusiform sinkers are used in gill nets. Spheroidal forms are rarely met with in nets. We find these sinkers in a certain kind of gill nets used for fishing on muddy ground with a loop of bamboo through the perforation in each sinker. In hand-lines, however, the spheroidal form of sinkers are much used.

Sinkers made of potsherds. They are often found in shell-mounds of the Kwanto district and are more or less oblong in form with ground notches at the narrow ends (Fig. 85). The edges of these sinkers are usually ground. Roughly speaking their weight is 15—60 g.

Nets and Twines.

As I have described before, some potteries may be taken as net-sinkers, so that there is little room to doubt that prehistoric fishermen in Japan used some kinds of nets for fishing. Moreover we find in shell-mounds skeletons of many small fish such as sardine (*Clupea melanosticta* Schlegel), anchovy (*Engraulis japonicus* Schlegel), horse-mackerel (*Caranx trachurus* L.), etc., and certain kinds of other animals, such as *Platycephalus*, *Kareius*, *Sepia*, shrimps, etc. which are difficult to catch by other means than nets. The kinds of nets used in prehistoric times would be landing nets, gill nets, drag seines, etc.

On potteries and potsherds we frequently find casts of parallel twines crossing each other and thus forming a mesh-work (Fig. 100). Some archaeologists consider these as casts of netting, but on close examination we find no knot in it. Moreover meshes are very irregular in size generally. In some specimens of potsherds casts of threads are simply parallel to each other at one part and pass over to a mesh-work at another part, while in other specimens casts of threads are much crowded together without order (Fig. 99).

The size and nature of twines used for lines or netting may be imagined from casts on potteries and potsherds, from grooves on harpoon-heads, hooks, sinkers, etc. or from calibres of perforations in these objects. The diameter of the perforation in small clay sinkers is 1.5—3.0 mm., but most clay sinkers have a larger perforation, 4—8 mm. in diameter. As some large sinkers have a large perforation, 10—15 mm. in diameter, we can imagine that the prehistoric people used thick ropes too in fishing. The diameter of the transverse hole in harpoon-heads is about 3 mm. The breadth of grooves on hooks is 1—2 mm. From casts on potteries we know that twines are usually made of two simple strands, twisted either to the left-hand side or to the right and we observe that they are made of fine textiles (Figs. 99, 100). Each strand has a diameter of about 0.5 mm. I have not yet found a twine made in prehistoric times of more than two strands,

Net-twines now used are generally composed of three strands, twisted to the left-hand side and each strand is often composed of two or more slender threads in turn. In gill-nets, however, two stranded twines or single twines are preferred. Twines used for line-fishing are usually 1—2 mm. in diameter and composed of two strands, twisted either to right or left; but thicker twines, 5—10 mm. in diameter, are composed of three strands.

Boats.

There is no doubt that the people of the Stone Age in Japan had also some sea-going boats, as they had inhabited Oshima at the mouth of Tokyo Bay, Miyatojima, and Tashirojima near Kinkwazan and as they caught some pelagic fish such as different kinds of bonitos, albacore, swordfish, etc.

An old dug-out boat of camphor-wood, kept in the Osaka Museum is very interesting and most remarkable in structure (Pl. XXVI). It was discovered in 1878 while opening a canal called Itachigawa at Namba, Osaka, imbedded in the soil about three and half metres under the surface. At the anterior part of the canoe a large portion of wood is rotten and worn away, so that the shape of the bow cannot be discerned well; but the general form of the canoe may more or less be understood as it seems that the length was not much reduced by wearing.

The boat is long lanceolate, shallow and is scoop-shaped at both extremities. The bottom of the boat is flat, more or less rounded and very thick. The breadth of the boat is contained more than eight times in its length and the depth more than twenty two times. From the measurement of the actual size, the boat is nearly 11.33 m. in length, 1.36 m. in breadth and about 0.50 m. in depth. The thickness of wood at the bottom is about 9 cm.

The remarkable point in the structure of the boat is that it consists of two pieces of wood, anterior and posterior, scarfed in a very peculiar way. These pieces of wood overlap each other for a distance of about 1.8 m. At this place, the upper half of the anterior piece of wood and

the lower half of the posterior piece are chiseled off with great care to fit each other tightly. These overlapping parts are fastened together by two transverse bars which penetrate them at the ends. These transverse bars or thwarts are kept in position or are supported by a longitudinal bar beneath them. The longitudinal bar is about 2.9 m. long and about 17 cm. in width and height, triangular in cross-section and is made immovable in its turn by means of two additional transverse bars near its extremities. Underneath the longitudinal bar there is a groove bent in the form of a rectangle, which corresponds just to the outline of the lower surface of the bar (Fig. 101). The additional bars are not found at present, but in sketches made by Prof. S. Tsuboi and Mr. N. Yamazaki separately, about fourteen years ago, the posterior bar is found in its position. Two pairs of shallow pits on the inner side of the boat, on a level with the holes penetrated by the principal transverse bars, represent the position of the additional bars. Moreover, there are four shallow grooves on the upper edge of the longitudinal bar, corresponding respectively to the two principal and two supplementary transverse bars.

Of the two pieces of wood, the anterior piece is much longer than the other. The former is not straight at the posterior margin, but it has a notch at the left side. The notch seems to have not been made intentionally. On the right-hand side of the posterior piece of wood the gunwale is pretty well preserved. It is rather thin and narrow, about 27 mm. in thickness and about 150 mm. in height. Here we find four irregularly elliptical holes, probably excavated artificially for a contrivance to propel the craft, like similar holes for passing rowlocks made of ropes in some boats used at present. These holes however are about 30 cm. apart and thus they are too near each other, if we take them as intended for rowlocks. Underneath the thin margin, and parallel to it, there is a ridge of 70 mm. in width and 15 mm. in thickness on the external side of the boat.

We cannot believe, that the boat was propelled by a sail or sails, as we find no trace for erecting a mast, but as the boat is comparatively large, it would have wanted a large crew.

Through the kindness of Mr. S. TOSHIA, I had the opportunity of examining a fine terra-cotta bowl dug out within a distance of about one metre from the boat at the same time and from the same depth with the latter and now owned by Mr. YAZAYEMON ONO, who was the builder of the Itachigawa canal. The bowl is shallow, undecorated, reddish in color and its substance is fine and thin. It is 145 mm. in diameter and 45 mm. in depth. Undoubtedly it belongs to the so-called Yayoishiki or intermediate pottery.*

I believe that the remarkable boat can with safety be considered to belong to the Stone Age, as there is no trace of a metallic substance in the boat. If the people who made the boat had some metals to use, they would have not taken so much pain in uniting two pieces of wood as I have described before. On the other hand, as it was found with intermediate pottery, it is probable that the dug-out would have been made at the end of the Stone Age.

A canoe of such peculiar structure is not found in any of the primitive types of boats now used in Japan, nor in any other country. However there is a record in a compilation of historical sketches† relating to the Province of Owari, that a similar boat was discovered in 1838 at Morokuwa village near Nagoya. The work contains a rude illustration of the scene of digging out the boat, with a short explanation. From the explanation we learn that the boat was found from the basin of a small river running through the village. It was made of camphor trees, connected together or scarfed antero-posteriorly at three places and there penetrated through by transverse bars. In a manuscript written at that time the boat is stated to have measured about 22 m. in length and nearly 2 m. in breadth. Thus this boat was very narrow too. According to these writings a wooden image, supposed to be buddhist, old coins, etc., were found simultaneously from the neighborhood of the canoe, but these objects and the canoe itself are not extant at present.

Another record of an ancient boat is found in a work about the

* MUNRO—Prehistoric Japan. pp. 293-307.

† OKADA and NOGUCHI—Owari Meishozuyō. Nagoya 1844.

Province of Echigo.* The record is very short and incomplete, but we can judge from it, that the boat was quite different from that in the Osaka Museum. The boat was discovered in 1833 on the bank of the Agano River at Shimoshin (formerly Shimoshinden) near Gosen, washed out by a flood from a depth of about 6 m. The length of the boat was about 6 m. and its shape differed from the boats then used. A remarkable point in structure was, that baleen or whalebone was used instead of metals to connect different pieces of wood. It is stated that those who investigated the material of the boat, could not determine what kind of wood it was, and the boat was considered at that time as a drift boat from a foreign country.

According to SNOW† Kurilsky Ainu construct boats by fastening gunwale, thwarts, etc., with whale sinews or whalebone fibres. However we cannot believe that the boat from the bank of Agano River was drifted from the Kurile Islands, as the oceanic current flows toward the north-east.

Besides these, there is a dug-out boat, kept in the Hachiman Shrine at Taiho village, Ibaraki-ken. According to Mr. KAWASUMI‡ the boat is very simple, 6.4 m. in length, 0.6 m. in breadth and 0.2 m. in depth. It was found at the bottom of a marsh in the village. Another similar boat is kept in the Natagiri Shrine, Nishizaki village, Chiba-ken. It is small, about 3 m. in length and 0.6 m. in breadth. It is told that the boat drifted to the shore of the village and the interior was painted red. Though these two boats are old we do not know whether they were made in prehistoric times or not.

A boat of a very old type is kept in a shinto-shrine at Mihonoseki, Shimane-ken. The shrine is dedicated to KOTOSHIRONUSHINOKAMI, son of OKUNINUSHINOKAMI, who was the ruler of the western part of Hondo before the reign of the present Imperial Family. The boat is known by the name of "morotabune" or "amanohatofune", and is said to have been made after the boat on which a messenger from our Imperial ancestor

* SUZUKI—Hokuyetsu Seppu. 1842.

† SNOW—In Forbidden Seas. London, 1910.

‡ KAWASUMI—Journ. Anthropol. Soc. Tokyo. Vol. XIII. 1898.

crossed over to Mihonoseki to see KOTOSHIRONUSHINOKAMI to treat with him about the abdication of supreme authority. When the boat at the shrine becomes rotten, a new boat is rebuilt, imitating faithfully the preceeding one, so that the present boat is believed to be the same in type as the boat of the ancient time.

The boat is flat and nearly rectangular at the gunwale, but the sides are swelled up beneath it. Two thwarts are found at the gunwale. From a record kept in the shrine we understand that the boat was formerly made by excavating camphorwood and was painted with red clay. The total length, or the distance between the verticals at both ends, is nearly 7 m., the breadth about 1 m., and the depth about 0.5 m. The boat is propelled by oars only. One large oar, 283 cm. long, is used for steering at the stern, and small oars, 177 cm. long, for rowing at the sides.

The oar has a cross-wise handle at the upper end.

A kind of a fishing canoe now in vogue in Nakanomi, a brackish lake by Mihonoseki, is nearly rectangular at the gunwale and is closely alike to the sacred boat, except the bow, which is nearly vertical in the boat.

The boat is known under the name of "Soriko."

Representations of Aquatic Animals.

As the prehistoric people in Japan seem to have not much indulged in pictorial art, there are very few objects which are intended to represent aquatic animals or which bear their figures.

A clay dish, representing a shell of *Halotis* (Fig. 102), discovered in the shell-mound of Shiizuka is a treasure in the collection of Mr. T. TAKASHIMA. It is 154 mm. long, 112 mm. broad, and 53 mm. high. The finish is rather neat and artistic, though the likeness is of course not exact. It bears ornaments of a characteristic pattern of our neolithic pottery on the thickened margin, representing the inner lip of the shell, and also on the external side between the margin and a ridge, intended for the outer angle of the shell. At the angle we find eleven protuber-

ances in a row. They are nearly of the same size and nearly equally distant from each other. Near the last protuberance and at the margin of the dish there is a notch, probably representing a half-made spiracle. A little below this notch we find a small hole about 5 mm. in diameter, the use of which we cannot make out. The interior as well as the exterior of the dish is nearly smooth. I was told that a similar dish was obtained from the shell-mound of Okadaira, Ibaraki-ken, and is in the possession of a certain gentleman.

Mr. TAKASHIMA has also a thin and large dish, probably representing a shell of *Anodonta* (Fig. 103). It is 235 mm. in the longer diameter, 155 mm. in the shorter diameter and about 50 mm. in depth. The inner side of the dish is smooth, while the outer is entirely filled with impressions of rough cloth and parallel circinal lines, convoluting either dextrally or sinisterly. At the middle of one longer side, there is an elevation which seems to represent the ear-shaped projection at the dorsal border. At the border of each narrower end, there is a small notch, the meaning of which is not known.

In a large lug of an earthen-ware, collected by Mr. TAKASHIMA, we find a design which might have been taken from a shark's head. The lug is triangularly prismatic and has a large perforation on each side. These perforations communicate with each other, and each of them is surrounded by a groove. Two of these perforations are elliptical and seem to represent the eyes, while the remaining one, which is larger, round and a little more remote from the distal end, seems to be the mouth.

Some small tubes made by cutting long bones of a large bird were collected by Prof. S. TSUBOI from a shell-mound at the mouth of Susuya, Karafuto. They bear many interesting designs engraved on them, and in one we find figures which are considered to have been intended for a scene of whale-hunting (Fig. 104). The bone tube is about 58 mm. long and about 16 mm. in diameter, and has a transverse band of a shallow groove of about 5 mm. in the middle. On each side of this groove there is a rude drawing, the one quite similar to the other, but opposite in direction. The drawing seems to represent a huge whale

harpooned from a boat manned by about eight men. A small boat-shaped figure anterior to and above the figure of the whale cannot be explained.

Utilization of Aquatic Animals Caught.

There is little doubt that aquatic animals caught were principally consumed as food. It is also probable that the prehistoric people in Japan have procured and used oils, sinews and hides or furs from marine mammals. Moreover, we find traces of utilization of the skeleton and other hard parts of aquatic vertebrates. Some rod-like or spatula-shaped instruments made of whale skeletons were found in shell-mounds. From the shell-mound of Yoyama Mr. TAKASHIMA obtained an epiphysis of a whale's vertebra, at the centre of which a large hole has been made. This might have been used as a stand for a kettle or jar. A large tooth of a sperm-whale with a hole at the root was discovered at Yoyama and a canine tooth of a seal, also perforated at the root, from Kuwagasaki. Baleen was found in place of metals in an ancient, most probably prehistoric boat, discovered at Shimoshin, Niigata-ken, as described above.

A piece of bone with three perforations and belonging to the lateral series of a turtle's dorsal shield was obtained from Yoyama and is in the possession of Mr. TAKASHIMA (Fig. 133). The fan-shaped group of tail-bones of a flat-fish, consisting of the last vertebra and hippural bones (Fig. 106) found in the same locality, and vertebrae of some Elasmobranchiate fishes were found coloured with hæmatite. These coloured bones were probably used as ornaments. The utilization of the tail spine of a sting ray for a missile implement is described on page 333.

Molluscan shells were used for many purposes in the prehistoric age too. Some shells, such as those of *Haliotis*, *Pecten*, *Cytheria*, etc., seem to have been used as dishes, pan, ladles, or receptacles, as they are still at present used for the same purposes. As there are earthenware dishes from shell-mounds, representing shells of *Haliotis* and *Anodonta*, their original shells have no doubt been used as dishes simultaneously. Shells of *Cytheria* from shell-mounds are frequently found to contain hæmatite.

From shell-mounds of different regions we frequently find very fine sickle-shaped or circular strips of molluscan shells. They are called shell-bracelets by MUNRO. Probably they had been used as ornaments. As these objects are found sometimes in large quantity, and as many of them are not quite finished, we can understand the process of manufacture pretty well (Figs. 143—145). At first the umbonal part is broken and an aperture is made. Then the aperture is gradually enlarged by chipping and at last a narrow strip of shell remains at the margin. Surfaces and rough edges of such strips are ground and a bracelet-like object is produced. For this purpose bivalves with thick shells, such as species of *Arca* and *Pectanculus* are selected. In the shell of these species we often find a small perforation at the umbonal region. The perforation is generally elliptical and its short diameter is 3—5 mm. These perforated shells are believed by some scholars to have been used as net-sinkers. In Akita-ken such perforated shells are still used for this purpose.

Stone Fish-knives and Sticks and Spatulas of Bone.

Stone implements, quite similar to the so-called fish-cutter from North America, were found in different localities. They are 100—200 mm. long and 50—60 mm. broad (Figs. 97, 98). They are well polished, generally straight at the back and curved at the cutting edge, which is tolerably sharp. Near the back we find two holes for fastening a wooden handle.

From shell-mounds of the northern district of Hondo we frequently find many pointed sticks, 60—160 mm. in length and made of the long bones of mammals. They are polished and rather sharply pointed at one end, but near the other end they remain generally crude or unfinished. Some of them are stained grayish with something like blood and grease. Besides these there are many spatulate objects made also of long bones and measuring 110—140 mm. in length and 12—20 mm. in breadth (Fig. 96). They are generally flat and rounded at one end.

The pointed sticks are conjectured by some archaeologists to have served for pricking flesh at the table, and the spatulate objects for opening

shell-fish. They might have served so, but the conjecture has not enough ground. Bent spatulate instruments are used at present for detaching ear-shells from rocks, but the bone spatulas are short and straight, so that they would be not good for this purpose. For opening bivalve shells, thin spatulate instruments are used now, but bone spatulas are thick at the margin. Moreover, as shells from shell-mounds are rarely broken at the margin, we are inclined to believe that in prehistoric times molluses were boiled before removing the flesh from the shell and the people of the time wanted no special instrument for shelling.

Remains of Animals in Shell-mounds.

Species of animals represented by their remains in shell-mounds differ very little from those now fished for; but there are some interesting changes in the distribution and abundance of animals when compared with their condition at present.

In a very large shell-mound of Yoshinomura near Kumamoto, on the coast of Yatsushiro Bay, shells of *Arca granosa* LINN. is most predominating, and other kinds of shell-fish, *Cyclina* and a few other kinds, are very insignificant in number. Nearly the same fact is found in a small shell-mound of Koama, also near Kumamoto but on the coast of Ariake Bay. Though these two bays are at present also very muddy and fit for the growth of *Arca granosa*, this species is very scarce, and is altogether valueless to the inhabitants round those bays, and *Tellina*, *Solecurtis* and *Ostrea* are extensively cultivated there. The size of shell, however, is larger at present as the mollusc is not crowded together.

We find bones of *Pagrus major* SCHLEGEL in a large quantity in shell-mounds at the northern coast of Tokyo Bay and in those round Kasumigaura, a brackish lake near the mouth of the Tone River. In waters near these localities, however, the fish is no more found at present.

It is also interesting to note that bones of big, old specimens of mackerel, horse-mackerel, *Monacanthus*, and *Tetrodon* are found abundantly in

shell-mounds of Miyagi-ken and Iwate-ken. In these districts these fishes are seldom fished for nowadays. Moreover bones of the sword-fish (*Tetrapturus?*) were found in shell-mounds of these districts, though the fish is not caught at present. *Pagrus cardinalis* LACÉPÈDE and *Sparus schlegeli* BLEEKER are frequently found in the shell-mound of Kuwagasaki, but both species are rarely met with in the market of the town at present.

There is no doubt that in prehistoric times aquatic animals were teeming prolifically in the littoral region and they were caught in waters nearer and shallower than the fishing grounds of the present day. Notwithstanding this, we are surprised to find the skeletons of *Gymnosarda pelamis* (LINNAEUS) rather abundantly in the shell-mound of Kuwagasaki, as this fish is found in the course of the warm current or Kuroshiwo, which runs 20—60 miles off shore in this district now. At first I thought that bones of this fish were of recent origin and mingled by chance among prehistoric remains, but by careful examination I found these bones also in deep, unmolested layers with other prehistoric objects, so that there is no doubt that this fish was caught by the prehistoric people too.

It is also surprising to find evidences that the prehistoric people in the northeastern part of Hondo caught fishes in rather deep sea, about 80 fathoms. Such fishery seems to have developed as the sea in this region is abruptly deep, the hundred-fathom line being reached in 5—10 mile's distance from the shore. Jaw-bones, opercular bones, etc. of certain species of the genus *Sebastes*, and spines of the dorsal fins of *Acanthias* sp. are often met with in shell-mounds. Only one vertebra of *Pterothrissus gissu* (HILGENDORF) was obtained by the author from the shell-mound of Kuwagasaki. The vertebra, 6 mm. in diameter, has a very large central perforation, the diameter of which is half the diameter of the vertebral centrum, and moreover there are five or six longitudinal ridges on each lateral side between the haemal and neural arches. These fishes are also caught now by long lines, and in the southern part of Hondo in deeper waters, 200 fathoms or more.

In shell-mounds many delicate structures such as scales, spines, fin-rays, etc., are often preserved in good condition. Crustacean shells are

seldom preserved in shell-mounds, but after a painstaking examination I found a few small fragments (Fig. 128).

By chance I found some vertebrae of *Engraulis japonicus* SCHLEGEL in mud attached to bones of a fox from the shell-mound of Yoyama. Since then I wash skeletons and other objects from shell-mounds in a basin and after taking off the large objects cleaned first, I repeatedly wash the sediment left until the turbidity becomes slight. After drying, the sediment is examined under a magnifying glass for small skeletons. Sometimes we collect a part of soil where minute skeletons are found in large quantity and treat it in the same way.

Though molluscan shells from shell-mounds are easily identified, remains of other animals are very difficult to identify. It is especially the case when their skeletons consist of many different parts, as those parts are found disconnected and even those disconnected parts seldom occur complete. Generally fish scales are difficult objects and insufficient for the identification of species.

It is noteworthy that certain bones are often and better preserved than others, so that we can imagine that certain kinds of fish would be better preserved than other species. When the number of individuals caught is large, the different kinds of bones found are also numerous. *Lateolabrax japonicus* is generally represented by its thick operculum and jaw bones; *Pagrus major* SCHLEGEL by jaw bones, palatine, coalesced frontals, occipital bones, etc.; *Pagrus cardinalis* LACEPEDE by occipital crest and jaw bones; *Sparus schlegeli* BLUKER by jaw bones; *Scomber colias* GMELIN and other scombroid fishes by vertebrae; *Caranx trachurus* L. by scutes, vertebrae and thick supraclavicle; flat fishes by the first interhaemal spine; *Mugil oeur* FORSKAL by thick operculum; *Monanthus* by the first dorsal spine and urohyal; *Tetrodon* by jaws; sharks and rays by teeth, spines and vertebrae.

Most of the animals sought after by the prehistoric people are the prime food-fishes of our country. Fishes commonly and abundantly found in shell-mounds are *Pagrus major*, *Sparus schlegeli*, *Lateolabrax japonicus* and *Thunnus thynnus*. It is noteworthy that remains of Tetrodontoid fish

are often found in shell-mounds. These fishes are poisonous and are not touched by wild animals. However these fish are much esteemed by gastronomers and the primitive people seem to have also appreciated the delicacy. As to the size of animals, there is a slight difference between those caught in the prehistoric time and those caught at present.

The following table gives a list of animals found in the shell-mounds of Japan and their distribution. Besides the animals enumerated in the table there are still many species which I cannot identify as yet. I have not much studied the molluscan shells from shell-mounds. Therefore I give here the names of shells published by Dr. A. Oka* with a few alterations and also with the addition of names of some shells collected by myself from Iwate-ken and Miyagi-ken.

* OKA—Shells from Japanese Shell Mounds. *Bullet. Tokyo. Anthropol. Soc.* Vol. X. 1895.

[illegible]

[illegible]

	Parts of the skeleton	Kuwagasaki	Ozuke	Osozawa	Nakazawa	Hosoura	Numazu	Yashikihama	Miyatojima	Yoyama	Shiizuka	Horinouchi	Sonno	Tachiki	Natagiri
<i>Pectunculus albolineatus</i>															
„ <i>fulguratus</i>															
<i>Cardium muticum</i>															
<i>Corbicula leana</i>															
<i>Venus jodoensis</i>						x		x							
<i>Cytheria meretrix</i>															
<i>Gomphia melanaegis</i> ?		x													
„ sp.		x													
<i>Unetia menstrualis</i>		x				x									
<i>Dosinia troscheli</i>		x													
<i>Cyclina chinensis</i>															
<i>Saxidomus purpuratus</i>								x							
<i>Tapes philippinarum</i>		x													
<i>Macra sachalinensis</i>															
„ <i>veneriformis</i>															
„ <i>sulcataria</i>															
<i>Tresus nuttali</i>					x										
<i>Solen gouldi</i>										x					
<i>Soletellina</i> sp.		x				x									
<i>Mya arenaria</i>															
„ <i>truncata</i>						x									
<i>Panopaea</i> sp.															
PISCES															
Elasmobranchii															
<i>Sharks, several specico</i>															
(<i>Lamna</i> , <i>Acanthias</i> , etc.)	Teeth, spines and vertebrae	x				x				x					x
<i>Trygon akajei</i>	Caudal spine	x						x	x	x					
<i>Myliobates tobije</i>	Teeth and caudal spine	x							x	x			x		

	Parts of the skeleton	Kuwagasaki	Ozuke	Osozawa	Nakazawa	Hosoura	Numazu	Yashikihama	Miyatojima	Yoyama	Shiizuka	Honnouchi	Sonno	Tachiki	Natagiri
Teleostei															
<i>Pterothrissus gissu</i>	Vertebra	x													
<i>Clupea melanosticta</i>	Vomer, jaw bones, vertebrae	x					x					x			
<i>Engraulis japonicus</i>	Vertebrae	x				x				x					
<i>Onchorhynchus</i> sp.	Vertebrae	x													
<i>Cyprinus carpio</i>	Hyomandibular, parietal, operculum, vertebrae, &c.													x	
<i>Leuciscus hakuensis</i>	Dentary	x													
<i>Anguilla japonica</i>	Dentary, hyoman- dibular, artic- ular, vertebrae, &c.											x			
<i>Mugil oeur</i>	Operculum, verte- brae									x		x		x	x
<i>Gadus brandti</i>	Vomer, jaw bones, otolith, verte- brae	x			x	x									
<i>Lateolabrax japonicus</i>	Jaw bones, oper- culum, preoper- culum, vertebrae	x	x	x	x		x			x				x	x
<i>Sebastes</i> sp.	Jaw bones, oper- culum, preoper- culum, etc.	x		x	x										
<i>Sparus schlegeli</i>	Jaw bones	x			x	x	x			x			x	x	
<i>Sparus aries</i>	Dentary											x			
<i>Pagrus major</i>	Jaw bones, front- als, occipitals, opercles, verte- brae, &c.	x	x		x	x	x	x		x	x		x	x	x
„ <i>cardinalis</i>	Jaw bones, occipi- tal crest	x			x	x									
Scaroid fishes	Pharyngeal bones														x
<i>Caranx trachurus</i>	Frontals, jaw bones, clavicle, scutes, etc.	x										x			
<i>Seriola quinqueradiata</i>	Dentary, vertebrae, etc.	x		x											x
<i>Scomber colias</i>	Dentary, hyoman- dibular, verte- brae	x					x								
<i>Auxis tapeinosoma</i>	Dentary, opercul- um, vertebrae	x													
<i>Gymnosarda pelamis</i>	Jaw bones, oper- culum, vertebrae	x													
<i>Thunnus thynnus</i>	Jaw bones, fin-rays, vertebrae, etc.	x		x	x	x	x	x		x					

	Parts of the skeleton	Kuwagasaki	Ozuke	Osozawa	Nakazawa	Hosoura	Numazu	Yashikihami	Miyatojima	Yoyama	Shiizuka	Hornouchi	Sonno	Tachiki	Natagiri
<i>Scomberomorus chinenses</i>	Dentary									x					
Sword-fish (<i>Tetrapturus</i> ?)	Jaw bones, vertebrae				x		x		x						
<i>Paralichthys olivaceus</i>	Jaw bones, hip-pural bones, vertebrae	x				x	x	x		x				x	
<i>Kareius bicoloratus</i>	Jaw bones									x					
Other flat-fish	First interhaemal spine	x			x							x			
<i>Platycephalus indicus</i>	Dentary, preoperculum, vertebrae											x	x		
<i>Monacanthus</i>	Premaxillary, spine, urohyal, vertebrae, etc.														
(two or more species)		x				x	x	x						x	
<i>Tetrodon</i>	Jaw bones, operculum, quadrate, vertebrae	x		x		x	x			x					
(several species)															
REPTILIA															
Chelonia															
<i>Caretta olivacea</i> ?	Hypoplastral plate									x					
MAMMALIA															
Cetacea															
Whales and dolphins	Teeth, otolith, vertebrae	x	x				x	x		x					x
<i>Physeter macrocephalus</i>															
<i>Orca gladiator</i>															
<i>Globiocephalus melas</i> , etc.															
Carnivora															
Seals	Teeth, phalangeal bones	x					x	x							

EXPLANATION OF PLATES.

PLATE XIX.

All figures drawn in actual size.

Figs. 1-6. Stone arrow-heads.

Fig. 7. A dart-head of stone, flaked. From Akita-ken.

Fig. 8. A dart-head of stone, ground. From Akita-ken.

Fig. 9. A flat arrow-head of bone or horn. From Hosoura.

Figs. 10-13. Conical arrow-heads of antler. From Miyagi-ken. (Fig. 11) and Iwate-ken.

Fig. 14. An arrow-nock of antler with coating of pitch at both ends. From Numazu, Miyagi-ken.

Fig. 15. An arrow-head of antler. From Togu, Miyagi-ken.

Fig. 16. A dart-head of antler with two barbs on one side. From Osozawa.

Fig. 17. A dart-head of antler, probably attached with its straight side to the end of a shaft. Transverse streaks of pitch showing the trace of lashing may be seen in the middle. From Miyatojima.

Fig. 18. A dart-head of antler with three unilateral barbs. From Osozawa.

Fig. 19. An arrow-head of antler with a pair of barbs. From Osozawa.

Fig. 20. A finely polished dart-head of bone.

Fig. 21. An arrow-head of boar's tusk. From Shiizuka.

Fig. 22, 23. Dart-heads of antler.

Fig. 24. A dart-head of antler like the one shown in Fig. 17. In this specimen a notch is found just above the place of lashing. From Miyatojima.

Figs. 25-27. Dart-heads of antler.

Fig. 28. A dart-head of the tail spine of a ray (*Trygon akajei*). From Yoyama.

PLATE XX.

Different kinds of nondetachable spear-heads or dart-heads. All figures drawn in actual size.

Fig. 29. A spear-head of antler from Mitsusawa.

Fig. 30. A dart-head or spear-head of antler with a constriction between the stem and the root. From

Fig. 31. A long spear-head of antler with five well defined barbs on one side and two indistinct barbs on the other. From Shiizuka.

Fig. 32. Largest spear-head of antler, broken at the distal portion. From Yoyama.

Fig. 33. A spear-head of antler with two barbs in two different planes. From Kamegaoka.

Fig. 34. A spear-head of antler with very numerous barbs and short root. From Osozawa.

Fig. 35. A peculiar form of a spear-head of antler with two constrictions, paired protuberances and pitch-coated root. Found at Osozawa.

Fig. 36. A dart-head or spear-head of bone. From Osozawa.

Fig. 37. A spear-head of antler with a perforation. From Osozawa.

PLATE XXI.

Different kinds of detachable harpoon-heads. All figures except Fig. 42 drawn in actual size.

Fig. 38. A harpoon-head of antler with the triangular root, which is broader than the stem. From Osozawa.

Fig. 39. A harpoon-head of antler with a perforated protuberance at one side. From Osozawa.

Fig. 40. A harpoon-head of antler with a constriction anterior to the short conical root. From Osozawa.

Fig. 41. A harpoon-head of antler with curved root. From Mitsusawa.

Fig. 42. A very long harpoon-head of antler with a lateral protuberance at the anterior end of the root. From Yoyama. $\frac{1}{2}$ actual size.

Fig. 43. A harpoon-head of antler with a perforation. The portion posterior to the perforation is missing. From Osozawa.

Fig. 44. A harpoon-head of antler with a perforation which runs in the plane of barbs. The apex and the portion posterior to the perforation are broken. From Numazu.

Fig. 45. A harpoon-head of antler with a perforation in the middle of the stem. From Miyatojima.

Fig. 46. A harpoon-head of antler with a constriction. From Miyatojima.

Fig. 47. A harpoon-head of antler with the curved tang, similar to the original of Fig. 41. From Miyatojima.

PLATE XXII.

All figures drawn in actual size.

Fig. 48. A prong of a fish-spear, made of antler. The distal end broken. From Miyatojima.

Fig. 49. A prong of a fish-spear with internal and external barbs and four external knobs at the proximal end. The distal portion broken. From Osozawa.

Fig. 50. A conical harpoon-head of antler with two spurs. From Miyatojima.

Fig. 51. A large prong of a fish-spear, with an internal barb. The distal end broken. From Miyatojima.

Fig. 52. A prong of a fish-spear with two internal barbs. The distal end broken. From Miyatojima.

Fig. 53. A conical harpoon-head with two barbs on one side and a spur on the other. From Osozawa.

Fig. 54. A long, conical harpoon-head, seen from the spurred side. The distal end of the spur is bifid, but one of its branches is broken. Faom Numazu, Miyagi-ken.

Fig. 55. A fine prong of a fish-spear, with a longitudinal groove at the internal side of the distal extremity. From Hosoura.

Fig. 56. A prong of fish-spear with an internal and two external barbs. There is a groove instead of a knob on the external side of the proximal end. From Miyatojima.

Fig. 57. A long, conical harpoon-head with a slightly bifid spur. From Yoyama.

Fig. 58. A harpoon-head with two nearly opposite barbs and two spurs. From Osozawa.

Fig. 59. A very complicated harpoon-head with a stone blade, three pairs of barbs and three spurs, one of which is broken. The apical portion of the stone blade is broken too. From Osozawa.

Fig. 60. A harpoon-head with two pairs of barbs and a spur. From Osozawa.

PLATE XXIII.

Hooks made of antler, drawn in actual size.

Fig. 61. A hook with an external barb, nearly in the base. From Kubiri, Kana-gawa-ken.

Fig. 62. A hook from Miyatojima. The apical portion broken.

Fig. 63. A hook with an internal and two external barbs. The tip of the internal barb is broken. From Osozawa.

Fig. 64. An angular hook with a curious barb. From Kashiwai, Chiba-ken.

Fig. 65. A thick hook with three external barbs. From Numazu, Miyagi-ken.

Fig. 66. A thick and rather clumsy hook with an internal and three external barbs. The upper extremity of the stem is broken. From Osozawa.

Fig. 67. A small hook with an internal and external barb. From Hosoura.

Fig. 68. A thick and large hook with an external barb and straight stem. From Yoyama.

Fig. 69. A small fine hook without a barb. From Osozawa.

Fig. 70. An angular hook with long stem. From Shiizuka.

Fig. 71. A small hook with an internal and three external barbs. From Osozawa.

Figs. 72, 73. Large finely curved hooks from Miyatojima. The apical portion is broken.

Fig. 74. A broken extremity of a hook with longitudinal groove. Seen from the internal side. From Yoyama.

Fig. 75. A small fine hook with two external barbs. From Osozawa.

PLATE XXIV.

All figures drawn in actual size.

Fig. 76. A landing gaff of antler. From Kamegaoka.

Fig. 77. A flattened clay sinker of dark greyish color with two crossing grooves. From Shiizuka.

Fig. 78. A more or less flattened elliptical clay-sinker with a groove round the edge. Partly reddish and partly greyish in color. From Shiizuka.

Fig. 79. A cylindrical clay sinker with a large perforation. Brownish in color. From Yoyoma.

Fig. 80. A more or less spherical clay sinker with three grooves crossing each other nearly at right angles.

Fig. 81. A flattened, elliptical stone sinker with two grooves. From Aizu, Fukushima-ken.

Fig. 82. A more or less fusiform clay sinker, brownish in color. From a place near Osozawa.

Fig. 83. A very small fusiform clay sinker, reddish in color. From Fukuda.

Fig. 84. A very large tubular clay sinker, purplish brown in color. From Fukuda.

Fig. 85. A sinker made of a potsherd.

Fig. 86. A nearly spherical clay sinker, with an unfinished margin of the perforation. From Fukuda.

Fig. 87. A clay sinker, similar to the original of Fig. 86, but the margin of the perforation is neatly finished. From Fukuda.

PLATE XXV.

All figures drawn in half actual size.

Fig. 88. A long stone sinker with two grooves. From Tomarioro, Karafuto.

Fig. 89. A flattened, elliptical stone sinker. From Onodai Akita-ken.

Fig. 90. An ellipsoidal stone sinker with a longitudinal groove. One extremity is broken. From Onodai.

Fig. 91. A flattened stone sinker with a longitudinal groove. From Nagai.

Fig. 92. A flattened stone sinker from Chichibu.

Fig. 93. A flattened stone sinker from Hobi, Atsumi Peninsula, Aichi-ken.

Fig. 94. A small flattened stone sinker from Aizu.

Fig. 95. A peach-shaped stone sinker with a longitudinal groove. From Tomarioro, Karafuto.

Fig. 96. A bone-spatula from Numazu, Miyagi-ken.

Fig. 97. A stone fish-knife from Miyagi-ken.

Fig. 98. A stone fish-knife from Miyagi-ken.

Fig. 99. A potsherd with casts of twine.

Fig. 100. A potsherd with casts of twines, crossing each other. The long twine on the left side is erroneously represented by the artist as twisted to the left. Kuwagasaki.

PLATE XXVI.

Fig. 101. A peculiar old boat in the Osaka Museum. The boat consists of two pieces of wood. The left end is the bow and the right end the prow. The third transverse bar is represented according to sketches of Prof. Tsuboi and Mr. Yamazaki.

Fig. 102. The same boat, seen from the anterior side. The posterior piece of the boat is lifted by interposition of a small piece of wood to show a groove beneath the longitudinal bar and internal depressions for the supplementary transverse bars.

PLATE XXVII.

All figures drawn in $\frac{3}{4}$ actual size.

Fig. 103. A clay dish, representing the shell of *Haliotis*. A portion of the external surface is fractured. The color is grayish brown. From Shiizuka.

Fig. 104. A clay dish, probably representing a valve of *Anodonta* shell. The color is grayish. From Yoyama.

Fig. 105. A piece of bird's bone with rude sketches of whale-hunting. From Karafuto.

PLATE XXVIII.

All figures drawn in actual size, unless otherwise stated.

Fig. 106. The last vertebra and hippural bones of *Paralichthys olivaceous* (Schlegel). Colored with red pigment. From Yoyama.

Fig. 107. Two scutes from the lateral line of *Caraux trachurus*. From Kuwagasaki. Twice the natural size.

Fig. 108. The operculum of *Lateolabrax japonicus* (Schlegel). Internal side. From Hosoura.

Fig. 109. An abdominal vertebra of *Tetrapturus* sp. From Nakazawa, Iwate-ken.

Fig. 110. A vertebra of an elasmobranch fish. From Yoyama.

Fig. 111. The third vertebra of *Gymnosarda pelamis*. From Kuwagasaki. The posterior end of the vertebra corresponds with the upper side in the figure.

Fig. 112. A vertebra of an elasmobranchiate fish. From Yoyama.

Fig. 113. The operculum of *Mugil oeur*.

Fig. 114. A vertebra of an elasmobranchiate fish. From Yoshino village, Kumamoto-ken.

Fig. 115. A large vertebra of an elasmobranchiate fish. From Hosoura.

Fig. 116. A tooth of *Myliobates tobijei*.

Fig. 117. A vertebra of *Scomber colias* (Gmelin). From Numazu, Miyagi-ken.

Fig. 118. The dentary of *Sparus aries*.

Fig. 119. The dentary of *Seriola quinqueradiata*. From Osozawa.

Fig. 120. Three vertebrae of *Engraulis japonicus* Schlegel. From Kuwagaki. Twice natural size.

- Fig. 121. The preoperculum of *Platycephalus indicus* L. From Sonno, Chiba-ken.
 Fig. 122. The premaxillary of *Sparus schlegeli*.
 Fig. 123. The preoperculum of *Sebastes* sp. From Nakazawa.
 Fig. 124. The dentary of *Tetrodon* sp. From Yoyama.
 Fig. 125. The first dorsal spine of *Monacanthus* sp. From Numazu, Miyagi-ken.
 Fig. 26. The coalesced frontals with the broken end of a dart head at the middle.
 Shiizuka.
 Fig. 127. The otolith of *Gadus brandti* Hilgendorf. From Nakazawa.
 Fig. 128. The rostrum of a shrimp. From Horinouchi. Twice natural size.
 Fig. 129. Two rays of a paired fin of *Thynnus thynnus*. From Osozawa.
 Fig. 130. The tail spine of *Miliobates tobiei*. From Miyatojima.
 Fig. 131. Pharyngeal of *Cyprinus carpio* L. From Tachiki.
 Fig. 132. The hyomandibular of *Anguilla japonica*. From Horinouchi. $1\frac{1}{2}$ natural size.

PLATE XXIX.

All figures drawn in natural size, unless otherwise stated.

- Fig. 133. A hypoplastral plate of *Caretta olivacea*? Three holes are drilled in it.
 From Yoyama. $\frac{2}{3}$ natural size.
 Fig. 134. The second vertebra of *Cyprinus carpio* Linneus. From Tachiki.
 Fig. 135. The dentary of *Scomberomorus sinensis* Lacep. From Yoyama.
 Fig. 136. A caudal vertebra of *Auxis tapeinosoma* Bleeker. From Kuwagasaki.
 Fig. 137. The dentary of *Kareius bicoloratus* Jordan and Snyder.
 Fig. 138. A vertebra of *Onchorhynchus* sp. From Kuwagasaki.
 Fig. 139. The cuttle of *Sepia*. From Tachiki.
 Fig. 140. Two vertebra of *Clupea melanosticta* Schlegel. From Kuwagasaki. Twice natural size.
 Fig. 141. Plates and spines of *Strongylocentrotus* sp. From Kuwagasaki. Twice natural size.
 Fig. 142. A canine tooth of a seal. From Kuwagasaki.
 Fig. 143-145. Series of designs illustrating the method of making shell-bracelets.
 $\frac{2}{3}$ natural size.

Description of the Clupeoid Fishes from Ogasawara or Bonin Islands.

BY

Kamakichi Kishinouye.

College of Agriculture, Komaba, Tokyo.

With Plate XXX.

After the publication of my paper* on our Clupeoids, I have received three kinds of fishes belonging to the same group from Ogasawara Islands. They are quite different from fellow-fishes, either of Hondo or of the Riūkiū Islands, and so far as I know they seem to be new to Science. Thus we find four regions in the distribution of the Clupeoid fishes in our waters—a northern region represented by the herring, the central region represented by *Clupea melanosticta* or *C. zunasi*, the southwestern or Riūkiū region represented by *C. mizun* and others, and lastly the southern or Ogasawara region.

Recently the fishing for bonito (*Gymnosarda pelamis*) has much developed round these islands, but one drawback is the deficiency of a suitable bait. The best bait is a clupeoid fish, but as the plankton is rather poor in the water round these islands, clupeoid fishes that feed on it are naturally rather small in their individual size as well as in the size of schools they make. The discovery of the three following species is the result of an ardent search for clupeoid fishes by bonito fishermen.

Clupea exile n. sp.

B. 6. D. 19. P. 14. V. 8. A. 16. Scales 44. 8.

Vertebrae 43. Cœc. pyl. ca. 60.

* KISHINOUE—Notes on the Natural History of the Sardine. Journ. Imp. Fish. Bureau, Vol. XIV. 1907.

Length of the head is nearly equal to the height of the body and is contained about $4\frac{1}{2}$ times in the total length. Diameter of the eye is equal to the length of the snout and is contained $3\frac{1}{2}$ times in the length of the head. Body is laterally compressed with the trenchant abdomen. Ventral profile more convex than the dorsal. Adipose eyelids present. No teeth found. Operculum smooth.

Origin of the dorsal fin is nearer the snout than the base of the caudal fin. Ventrals inserted a little behind the vertical from the origin of the dorsal. Anal fin is moderate in length and originates behind the vertical from the tip of the last ray of the dorsal. Caudal fin is covered with many minute scales. Scales rather adherent. Eighteen abdominal scutes before and fourteen behind the base of ventrals. All scutes consists of three pieces.

Alimentary canal has a little bend at the duodenum. Peritoneum black.

In the preserved specimens the dorsal and caudal fins and the dorsal part of the body dark greyish.

It is reported that this species reaches a length of about 15 cm. and is found all the year round, but especially abundant in autumn. Spawning of this fish is said to take place in spring. This is the most abundant clupeoid fish from Ogasawara Islands.

Vulgar name is "hon-iwashi" or true sardine.

Caught at Chichijima.

Clupea oguro n. sp.

B. 6. D. 13. P. 13. V. 8. A. 18. Scales 40, 11.

Vertebrae 40. Cœc. pyl. 35.

Length of the head is nearly equal to the height of the body and is contained about 5 times in the total length. Diameter of the eye is nearly $1\frac{1}{2}$ times contained in the snout and 4 times in the length of the head. Body is laterally compressed. Ventral profile is more convex than the dorsal. Adipose eyelids present. No teeth present. Operculum smooth.

Origin of the dorsal fin is nearer the snout than the base of the

caudal. Origin of the ventrals is just below the middle of the dorsal. Anal is about $11\frac{1}{2}$ times longer than the dorsal at the line of insertion. Minute scales cover the greater part of the caudal. Sixteen scutes before and twelve scutes behind the ventrals. All scutes consist of three pieces.

Alimentary canal has a little bend at the duodenum. Pyloric cœca are arranged in nearly three rows. Peritoneum black.

Back greyish. Tip of the upper and lower jaws and that of the lobes of the caudal are black.

Specimens before me may be divided into two groups, according to their size—5 cm. and 10 cm.

This species is caught together with the preceding species and is also very abundant in autumn.

Vulgar name is "oguro-iwashi" or black tailed sardine.

Our specimens were caught at Futami Harbour, Chichijima.

Engraulis macrops n. sp.

B. 11. D. 12. P. 11. V. 7. A. 32. Scales 40, 8.

Vertebrae 40. Cœc. pyl. 13.

Length of the head is contained about 5 times in the total length, and the depth of the body $5\frac{1}{2}$ times. Diameter of the eye is nearly equal to the length of the snout and is contained nearly 4 times in the head. Maxillary long and reaching the lower angle of the preoperculum. Very minute teeth on vomer, palatines, pterygoids and jaws. Body compressed and belly trenchant. Eyes subcutaneous. Snout rather blunt. Origin of the dorsal just in the midway between the snout and the base of the caudal. Caudal is deeply forked and is covered with minute scales. Pectorals scarcely reach the base of ventrals. Length of the anal is equal to that of the head. Scales with six to eight transverse striæ. Six scutes before and seven scutes behind the ventrals. Stomach black, peritoneum colorless. Intestine nearly straight. Back and dorsal and caudal fins greyish.

Type specimens measure about 12 cm. They were found in a school of *Caranx muroadi*, caught with a circle net at Hahajima in April, 1907.

EXPLANATION OF PLATE XXX.

All figures drawn in natural size.

Fig. 1. *Clupea exile*.

Fig. 2. *Clupea oguro*.

Fig. 3. *Engraulis macrops*.

Vol. II. No. 7, PUBLISHED DEC. 26th, 1911.

Price in Tokyo Yen 0.86

THIS JOURNAL IS ON SALE AT

Z. P. Maruya & Co., L'td.

Tori Sancho, Nihonbashi, Tokyo.

Geiser & Gilbert

Kaji-cho 23, Tokyo; Main Street 77, Yokohama.

Josephinenstr. 6. Leipzig.—R., Germany.

編纂兼發行者

東京帝國大學

明治四十四年十二月二十四日印刷
明治四十四年十二月二十六日發行

印刷者

中西美重藏

東京市下谷區谷中初音町四丁目拾七番地

印刷所

シヤパン、タイムス社

東京市麹町區內幸町一丁目五番地

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NOTICE.

VOL. I.

- No. 1, published June 16th, 1909.
- No. 2, published June 29th, 1909.
- No. 3, published March 28th, 1911.
- No. 4, in press.

VOL. II.

- No. 1, published December 8th, 1909.
- No. 2, published February 12th, 1910.
- No. 3, published April 30th, 1910.
- No. 4, published July 18th, 1910.
- No. 5, published October 31st, 1910.
- No. 6, published April 25th, 1911.
- No. 7, published December 26th, 1911.

VOL. III.

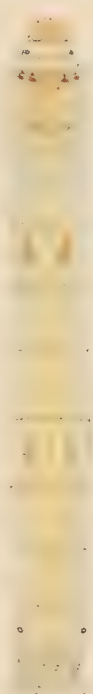
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 - No. 2, in press.
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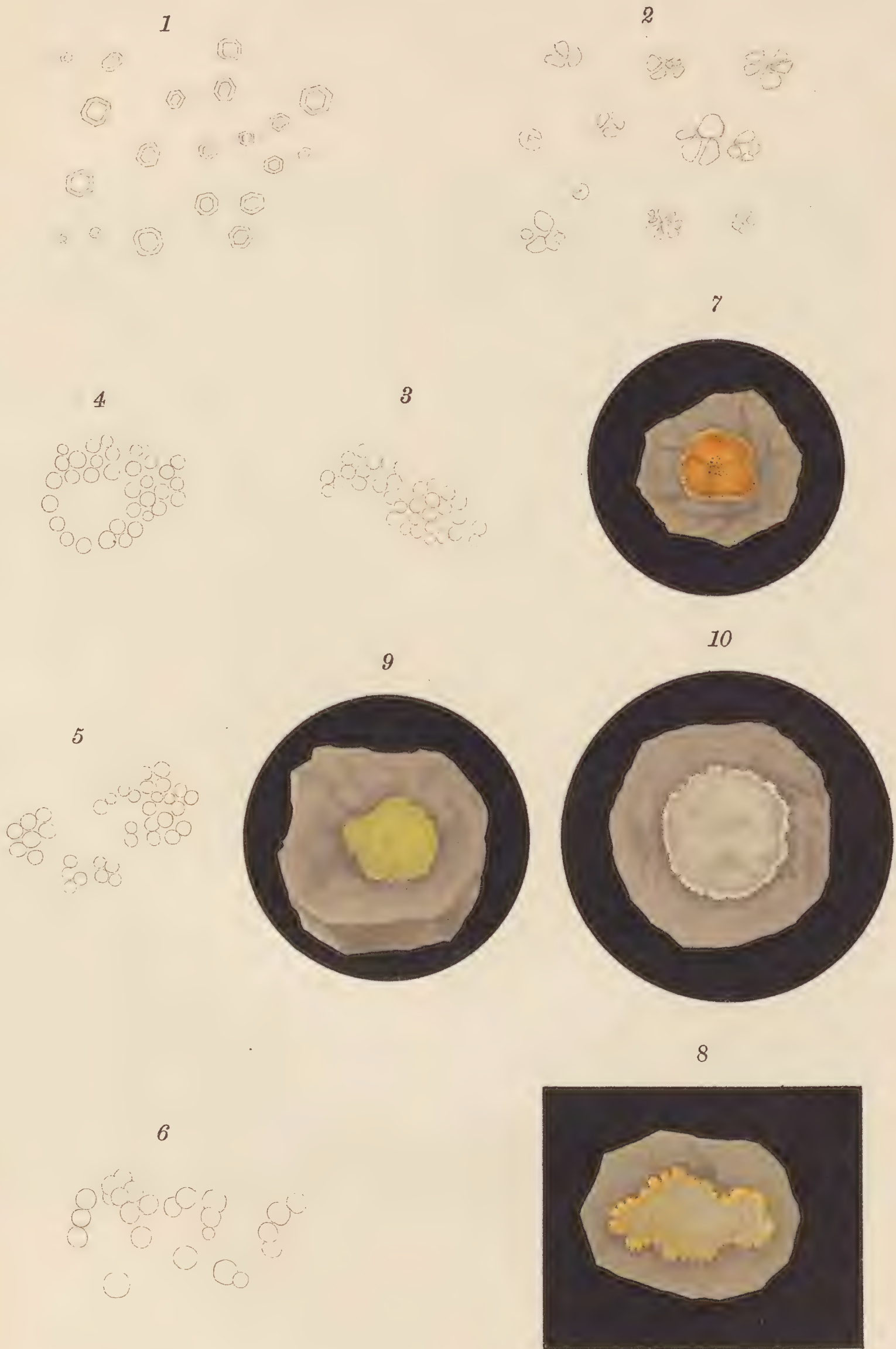
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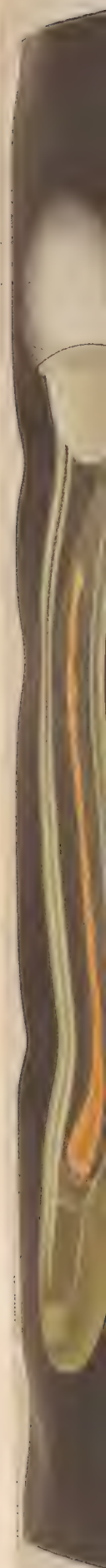


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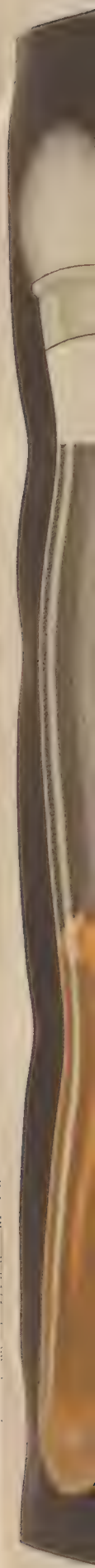


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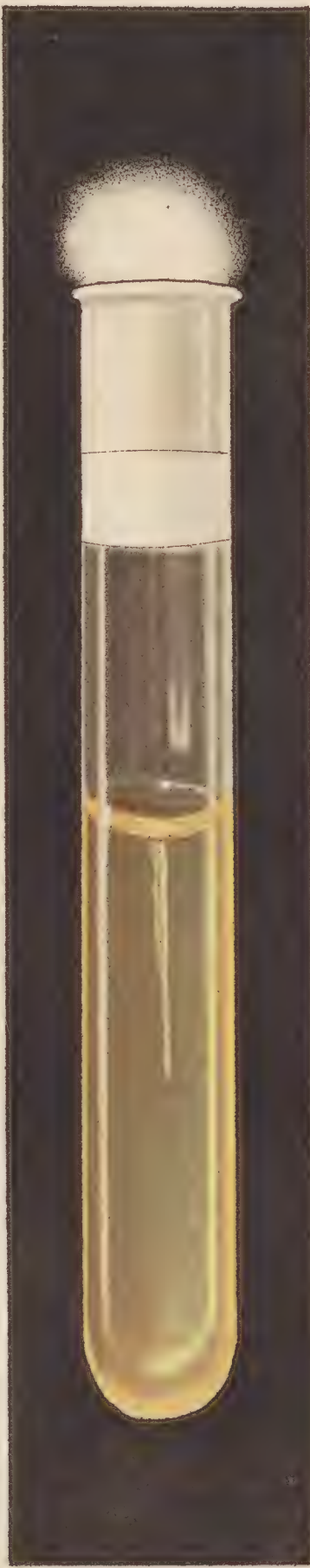


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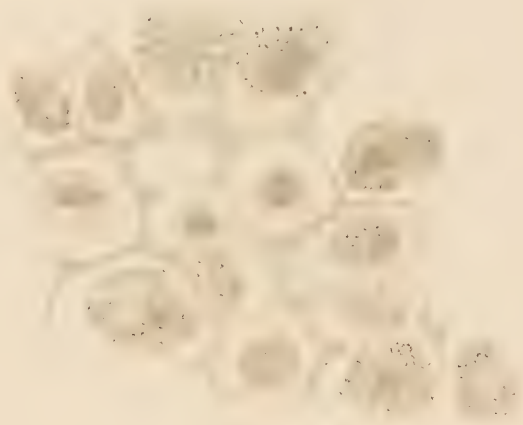
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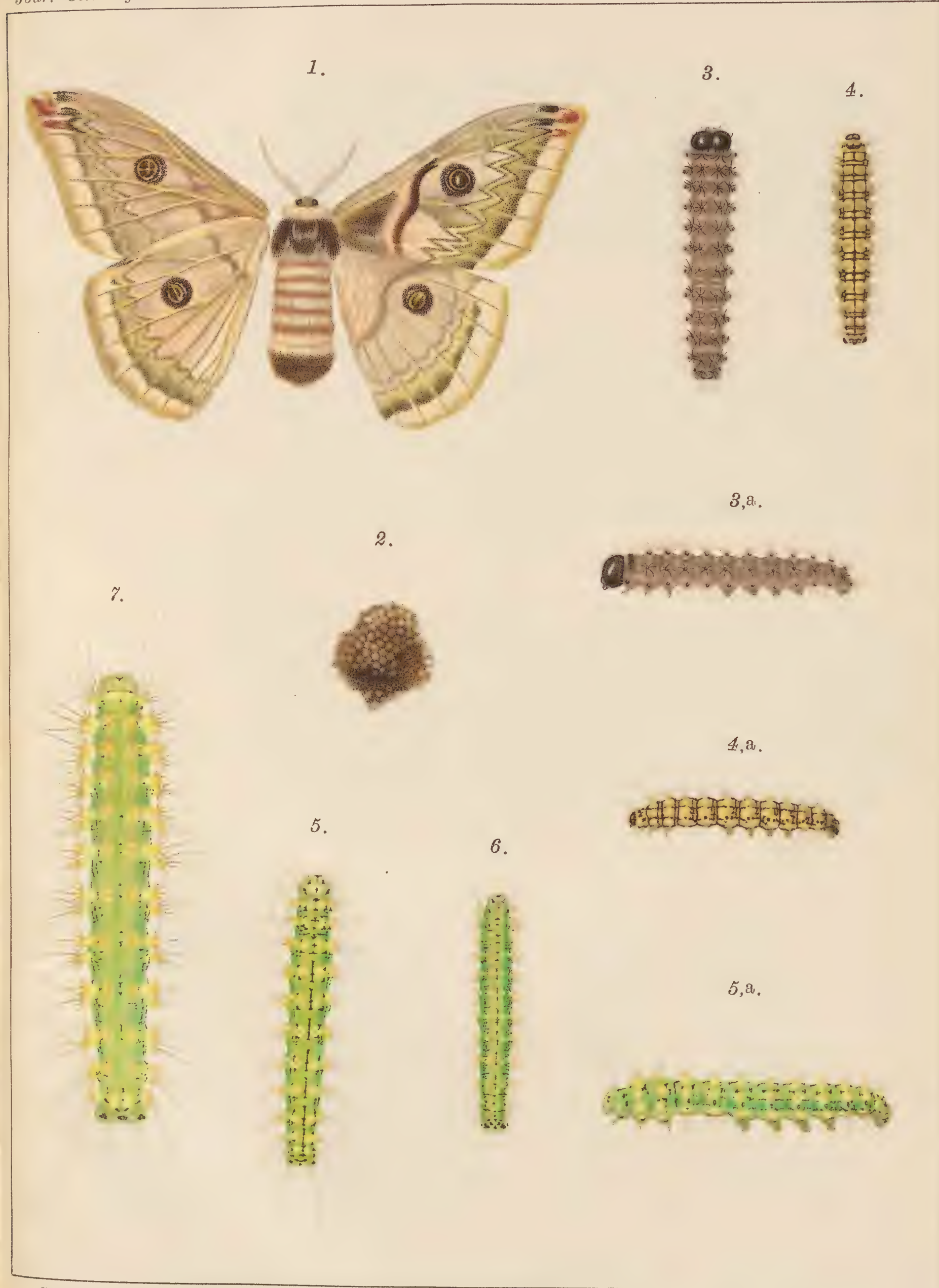
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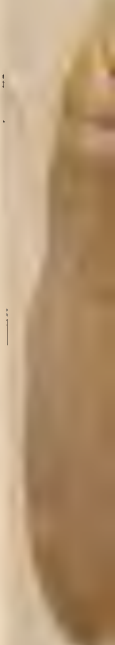


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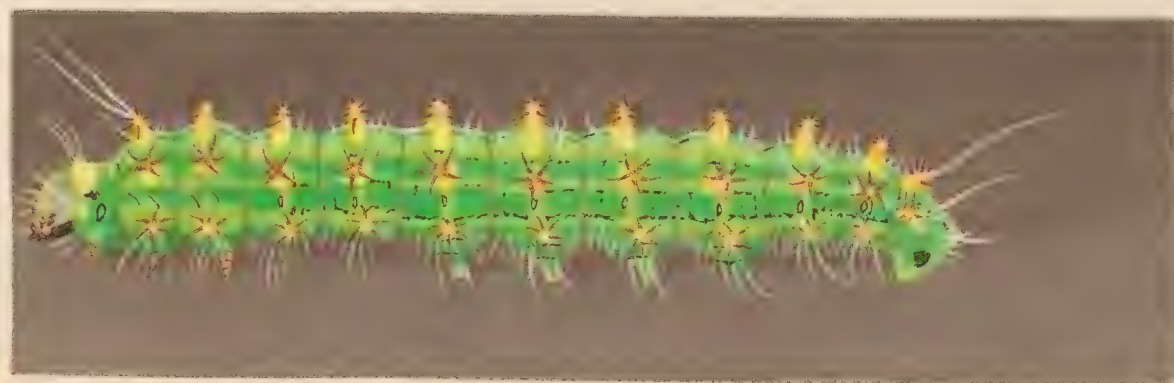




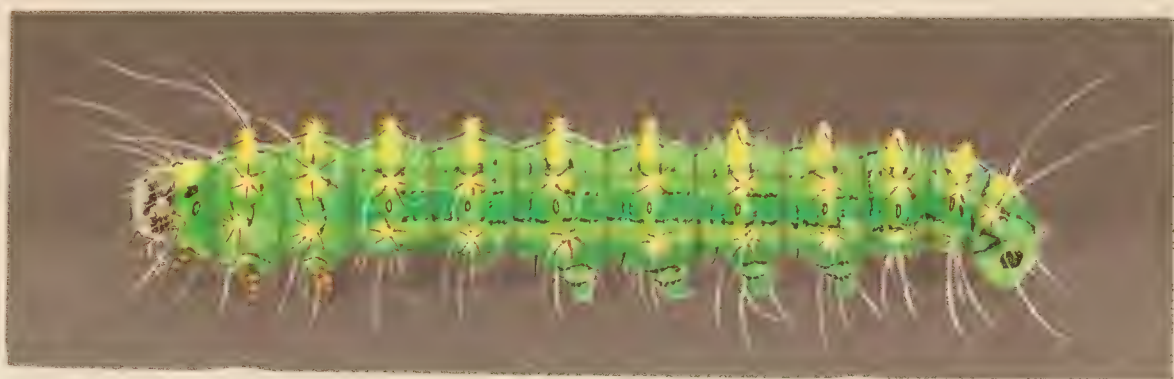


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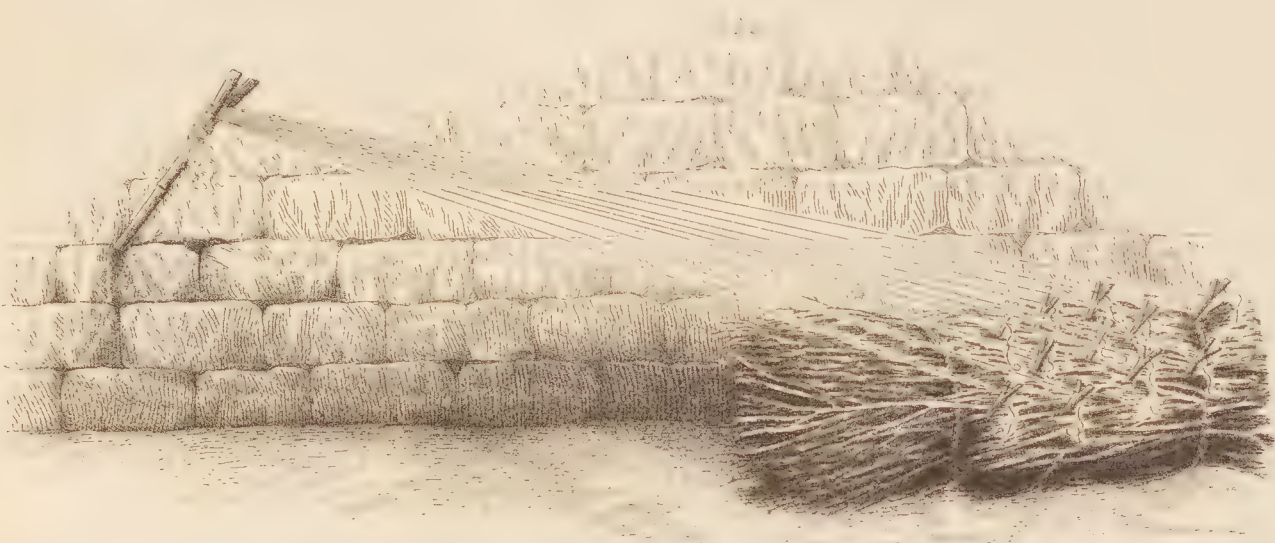




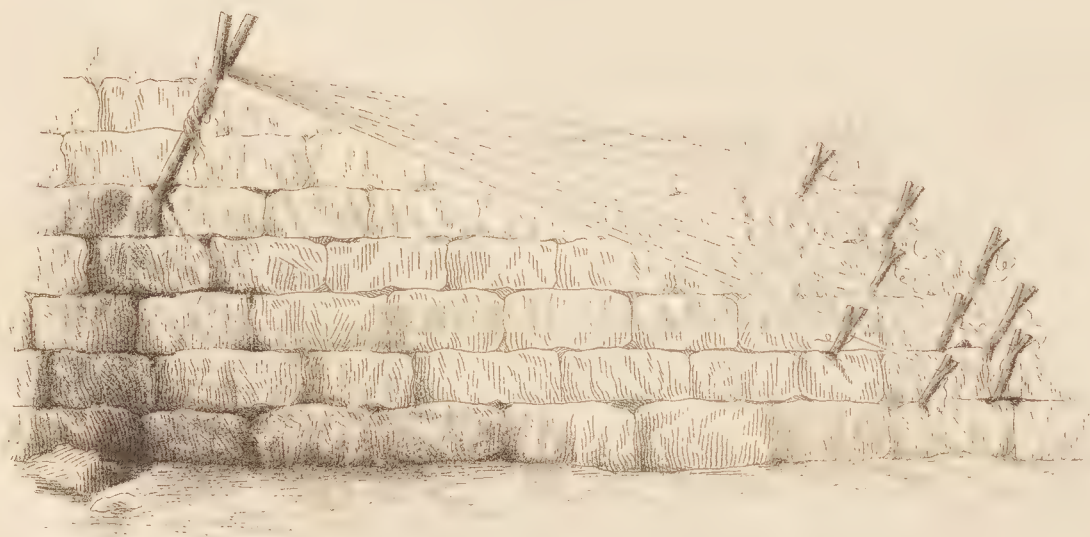
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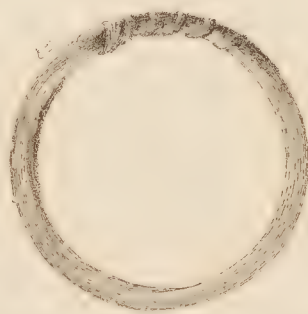
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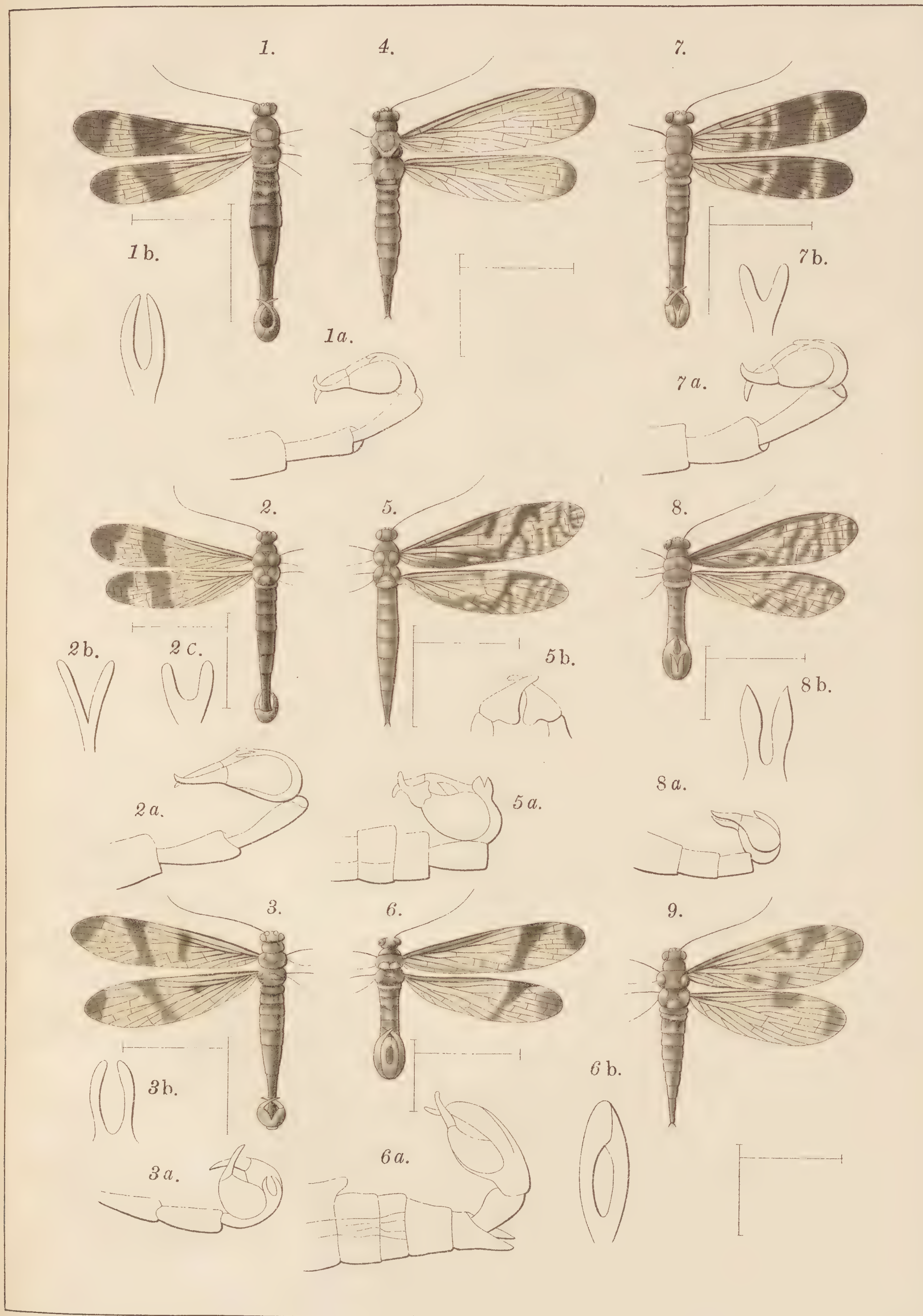


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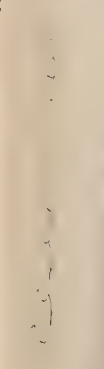
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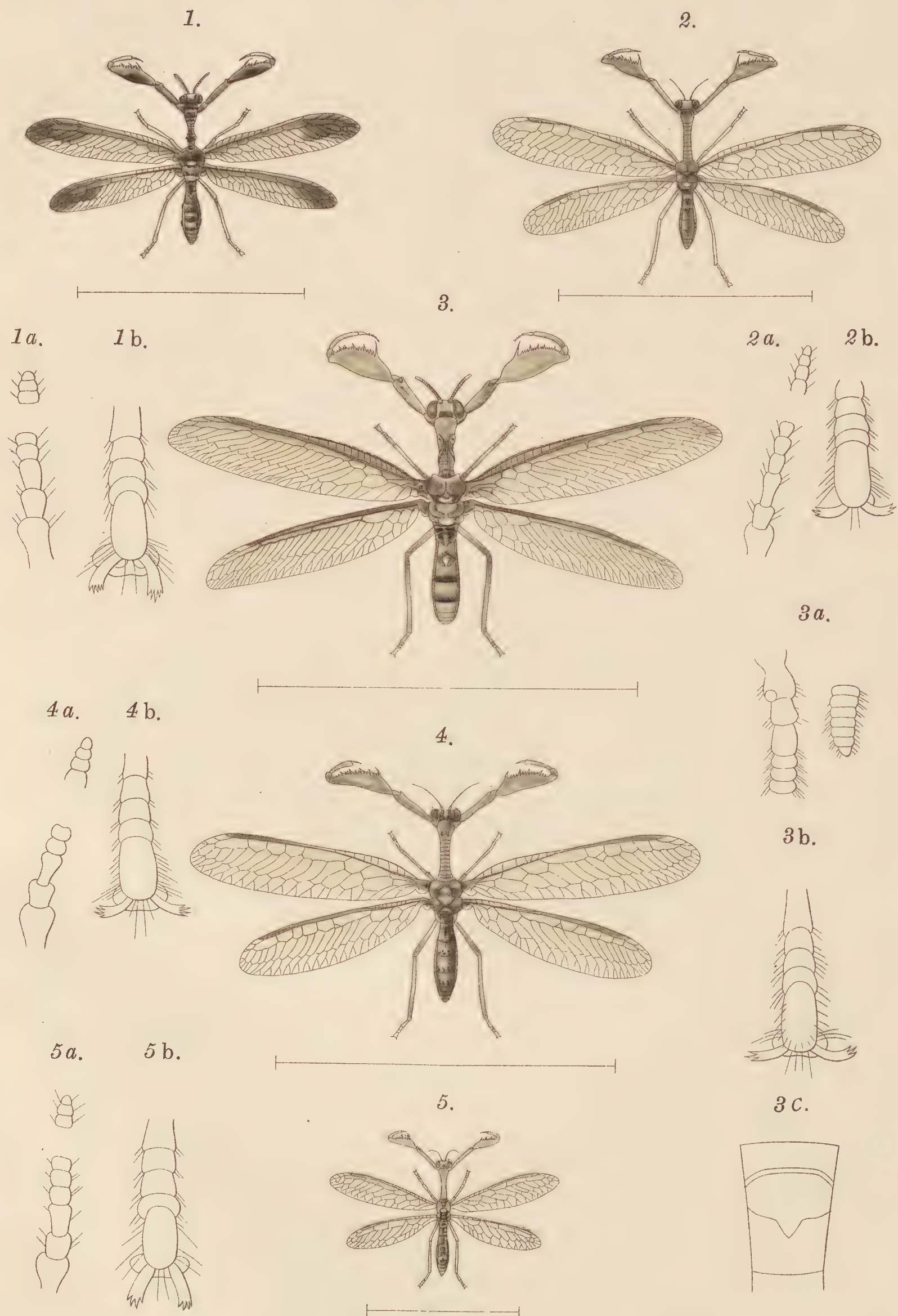


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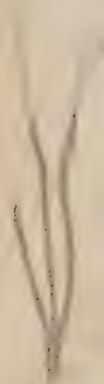


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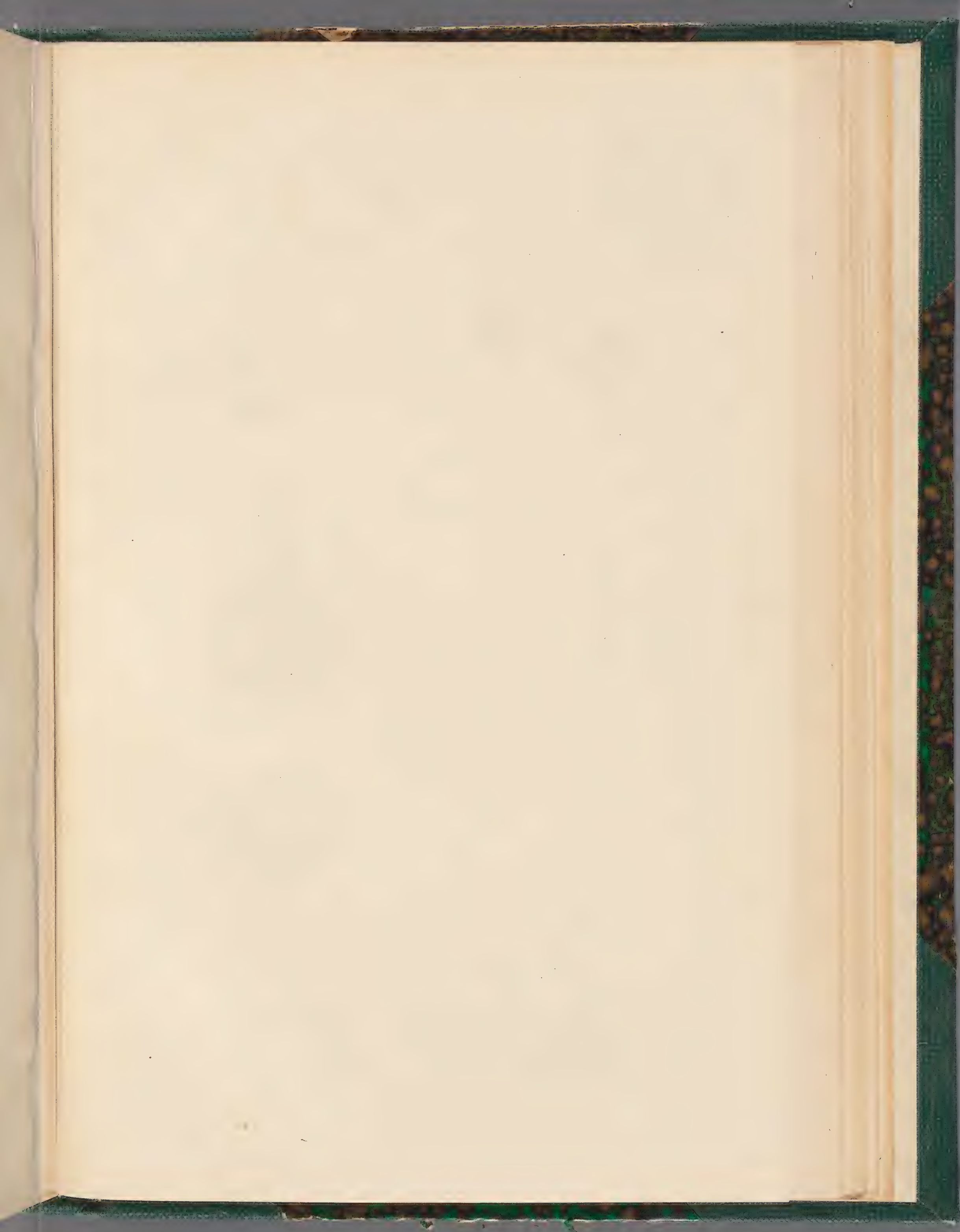


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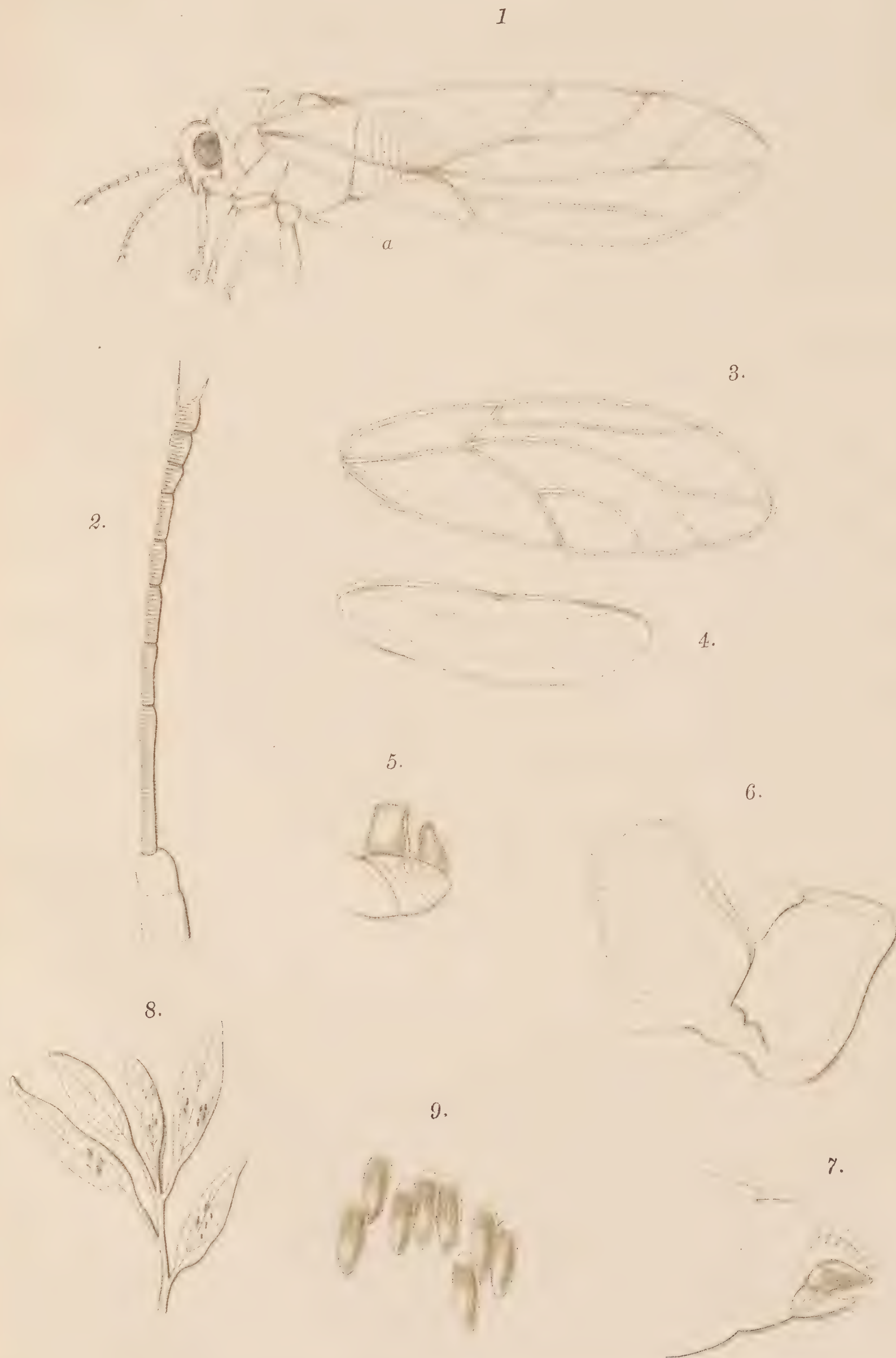


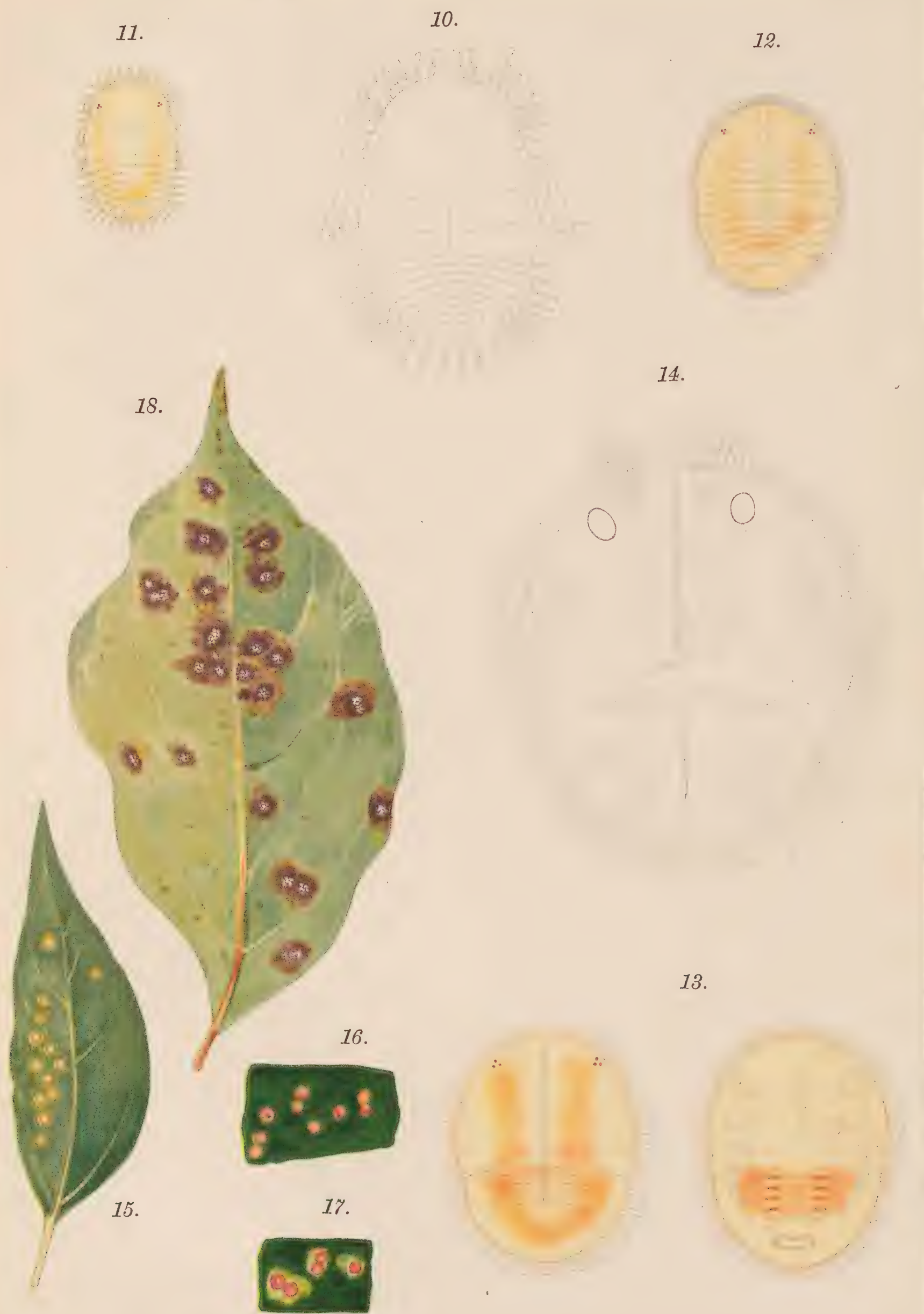
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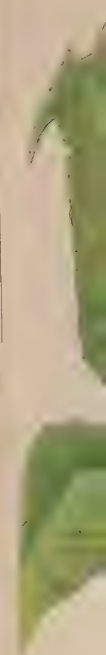








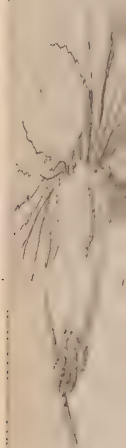






Kusano et Shiihara del.

Lith. H. Oye Tokyo.



19.



30.



41.



42.

43.







S. Kikkawa del.

Figs. 1—28.

H. Oye. imp.



S. Kikkawa del.

Figs. 29—37.

H. Oya. imp.



S. Kilkawa del.

Figs 38—47.

II. Oye. imp.



S. Kikkawa del.

Figs. 48—60.

H. Oye, imp.



S. Kikkawa del.

Figs. 61—75.

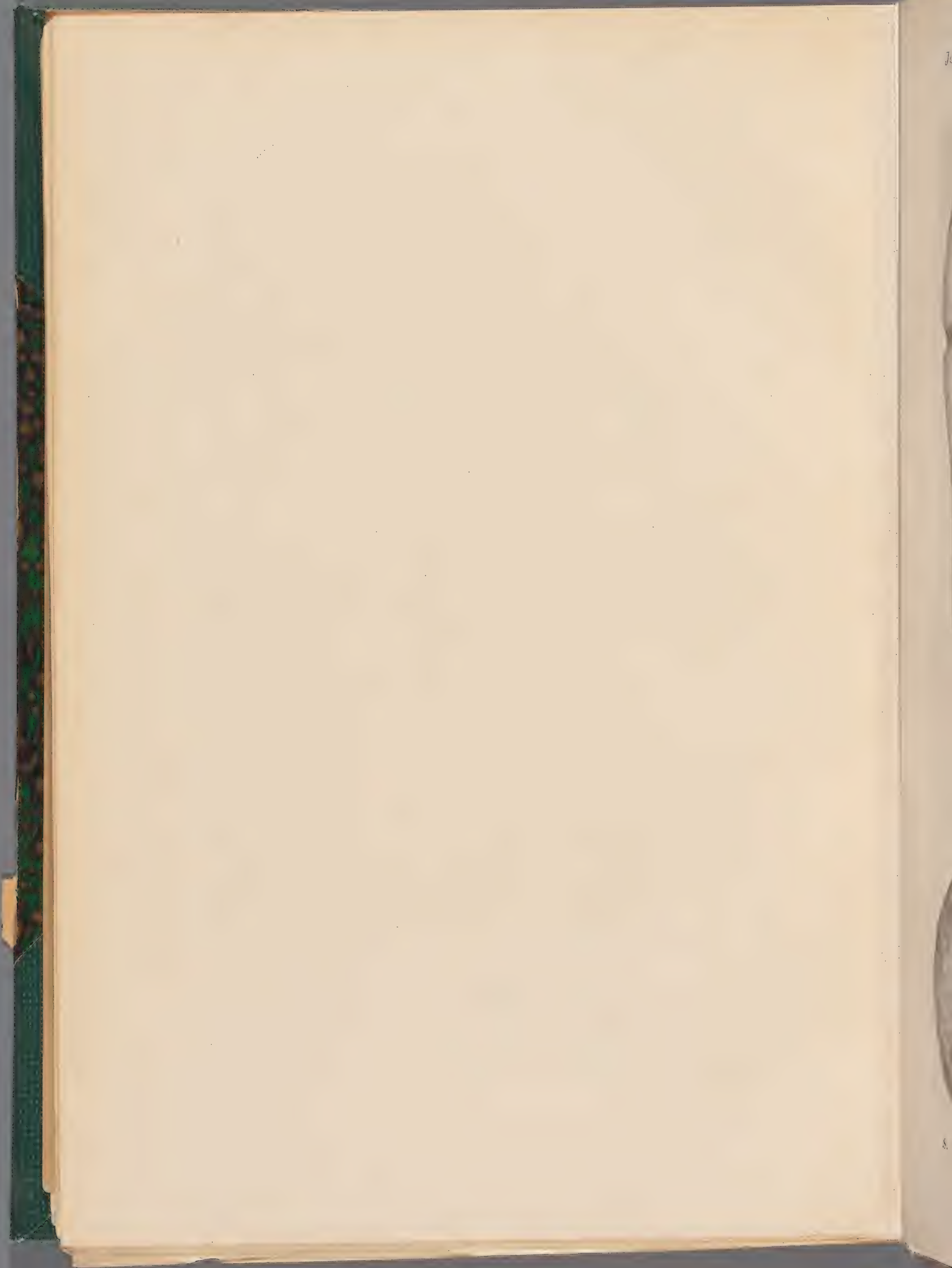
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Figs. 76—87.

H. Oye. imp.





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Figs. 88—100.

II. Oye. imp.

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1M.

102



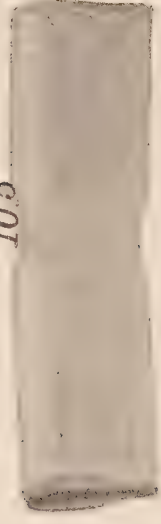
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H. Oyle. imp.

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S. Kikkawa del.

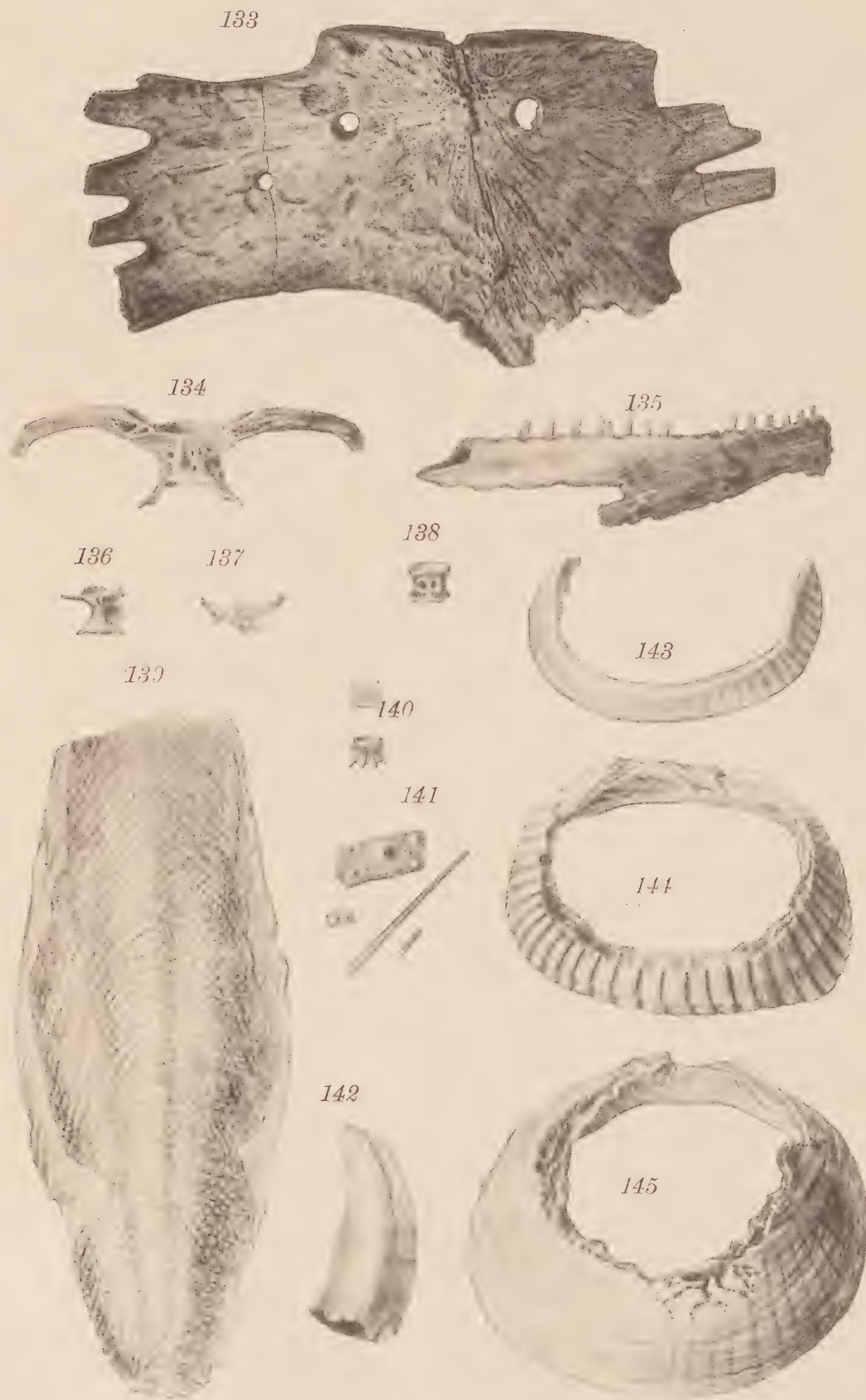
H. Oye. imp.

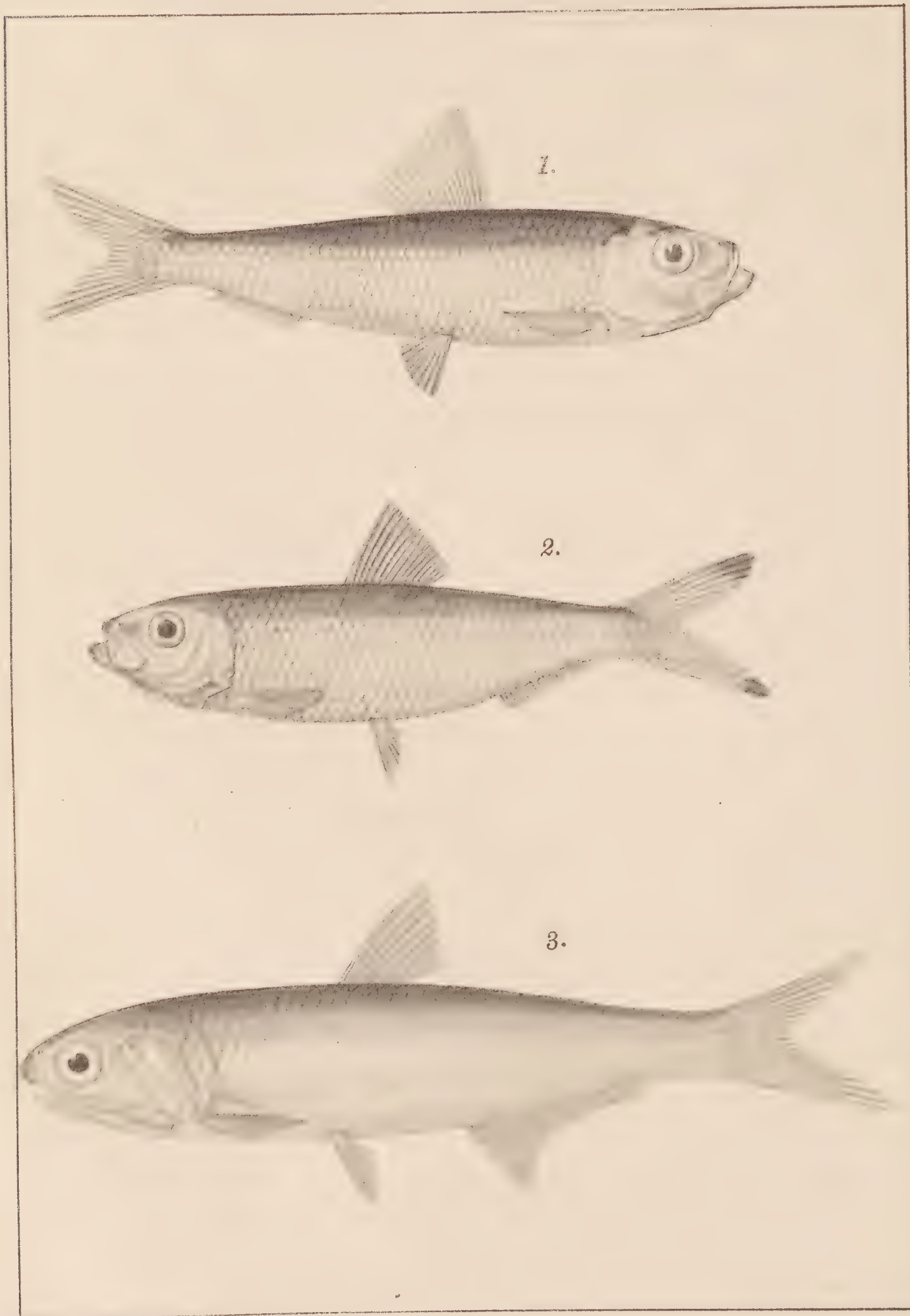


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Figs. 106—132.

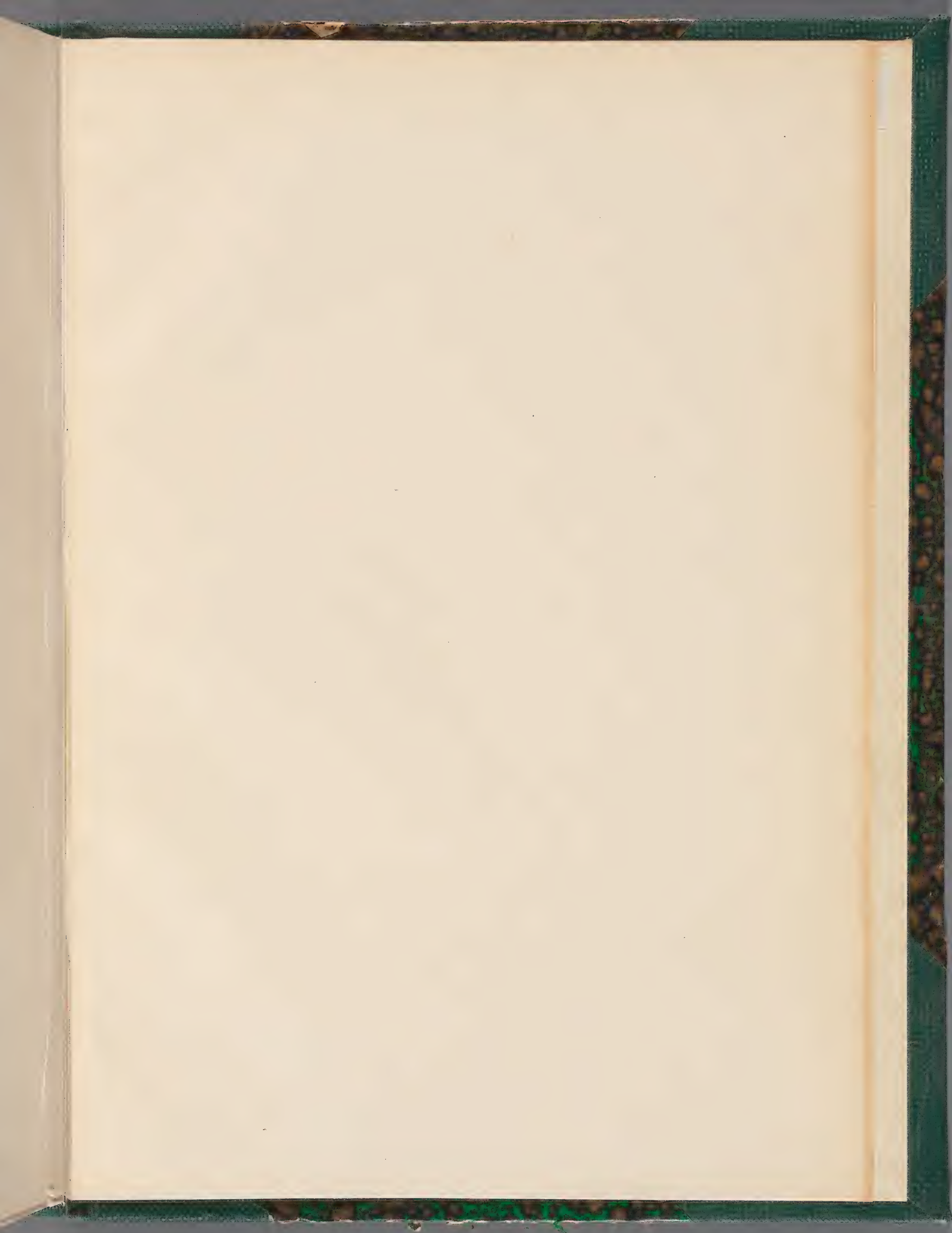
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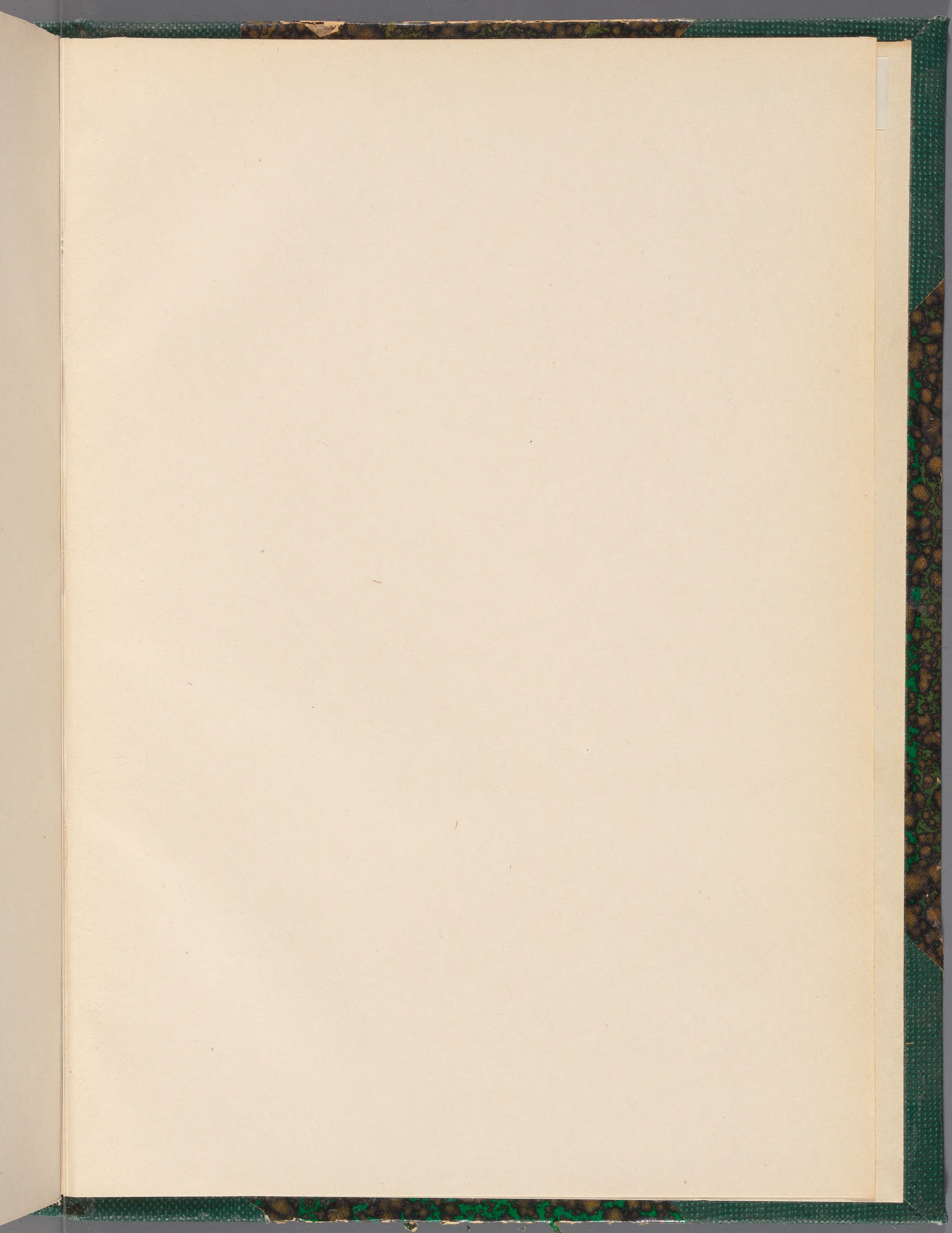




S. Kikkawa del. 1. *Clupea exile* 2. *Clupea oguro*. 3. *Engraulis macropus*.

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